The breeding season of pubertal red deer hinds

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ABSTRACT

In a study of seasonality of reproduction, 4 pubertal red deer hinds were monitored for live weight and plasma hormone concentrations from December 1987 to October 1988 (i.e. 12-22 months of age). On 7 occasions blood samples were collected every 20 minutes for 4 h to monitor secretion of luteinising hormone (LH) and response to an i.v. injection of 2 μg gonadotrophin releasing hormone (GnRH). Plasma progesterone profiles indicated that 4-6 ovarian cycles, lasting about 19 days each, occurred for each hind. Regular ovarian cycles commenced in late April (26 April ± 3.4 d, mean ± S.E.M.) and ceased about 3 months later. The number of LH pulses/4 h was higher in June than during the non-breeding season, but there was no change in the LH response to GnRH. These data showed that the breeding season of non-pregnant yearling red deer hinds was shorter than that reported for older hinds. Constancy of pituitary responsiveness to GnRH indicated that, as in other species, breeding seasonality is determined by changes in the output of GnRH from the hypothalamus rather than seasonal changes in sensitivity to GnRH.

Keywords Red deer hinds, puberty, breeding season, progesterone, LH, live weight.

INTRODUCTION

Red deer hinds exhibit highly seasonal changes in reproductive activity. The breeding season begins in autumn and, in the absence of pregnancy, adult red deer hinds may undergo 6-9 ovulatory cycles over a period of 5 months, with reproductive activity ending in September/October (equivalent to March/April in the Northern Hemisphere) (Adam *et al.*, 1989; Loudon *et al.*, 1989; Meikle *et al.*, 1991). In pubertal hinds the onset of the breeding season is influenced not only by photoperiod (Webster & Barrell, 1985) but by a requirement to attain sufficient body size before the transition to sexual maturity will occur (Clutton-Brock *et al.*, 1982; Fisher & Fennessy, 1985; Hamilton & Blaxter, 1980). Generally, farmed red deer hinds reach the threshold live-weight range for puberty of 65-70 kg before their second summer and a high percentage of yearlings conceive at about 16 months of age (Kelly & Moore, 1977; Fisher & Fennessy, 1985). Little is known of the duration of the first breeding season of pubertal red deer hinds in New Zealand or of related changes in patterns of hormone secretion during this period. In this study plasma progesterone concentrations were measured to monitor ovarian activity in pubertal red deer hinds throughout the breeding period and changes in reproductive activity were related to changes in the pattern of plasma LH secretion and pituitary responsiveness to GnRH.

MATERIALS AND METHODS

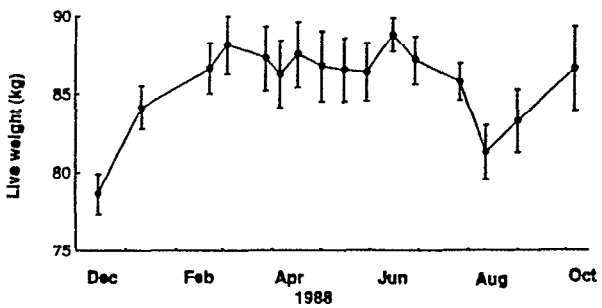
Four yearling red deer hinds were grazed on ryegrass/white clover pasture at the Lincoln University Deer Unit from December 1987 to October 1988 (i.e. 12-22 months of age). Infrequent blood samples were collected by jugular venepuncture at approximately fortnightly intervals from December 1987 to March 1988 and 2-3 times a week from April to October 1988 and the plasma analysed for progesterone and LH concentrations. On 7 occasions (15 December 1987, 29 February, 15 March, 24 April, 14 June, 29 June and 18 September 1988) jugular cannulation was performed and blood samples were collected every 20 min for 4 h to establish LH secretion patterns. Immediately following this intensive blood sampling 2 μg GnRH (1 μg GnRH/ml sterile 0.9% w/v saline solution, LH-RH/FSH-RH (amide form), NIAMDD, Lot 26-306 AL) was administered via i.v. infusion through the cannula and blood samples were collected after 10, 30, 60, 90, 120, 150, 180, 210 and 240 min. Plasma progesterone and LH concentrations were assayed using methods described by Duckworth & Barrell (1991). The sensitivity of the LH assay, calculated as 2 standard deviations from zero, was 0.22 ng/ml. Plasma samples with mean LH concentrations between 1.0 and 9.6 ng/ml had within- and between-assay coefficients of variation (c.v.) of less than 9% and 10%, respectively. The sensitivity of the progesterone ELISA was 0.23 ng/ml and plasma samples containing 0.7-6.2 ng/ml had within- and between-assay c.v. of between 9-13% and 8-14%, respectively. A vasectomised stag was run with the hinds from January until September 1988. Hinds (n=28) from the same age cohort, which were not blood sampled, were run with an entire red deer stag from 1 March and their calving dates recorded.

Results are expressed as mean ± S.E.M.. For each hind the onset of the breeding season was defined as the date when a luteal phase progesterone concentration (> 1 ng/ml > 5 d) was first recorded. Similarly, the onset of seasonal anoestrus was defined as the date when plasma progesterone concentration fell below 1 ng/ml following the last luteal phase of the season. An increment in LH concentration during an intensive blood sampling period was termed as a pulse if the increase (pulse amplitude) was more than 3 times the standard deviation of the previous sample. Mean LH was the average plasma LH concentration recorded during the 4 h intensive blood sampling period The LH response to exogenous GnRH was the difference between plasma LH concentrations immediately before and 10 minutes after GnRH administration. Live weight, progesterone and LH data were transformed to their logarithms (base ten) prior to statistical analysis. Data were analysed by ANOVA (SAS, SAS Institute Inc, USA) in conjunction with Duncan's LSD test to separate means when time was a significant factor. Significant differences indicate $p < 0.05$.

RESULTS

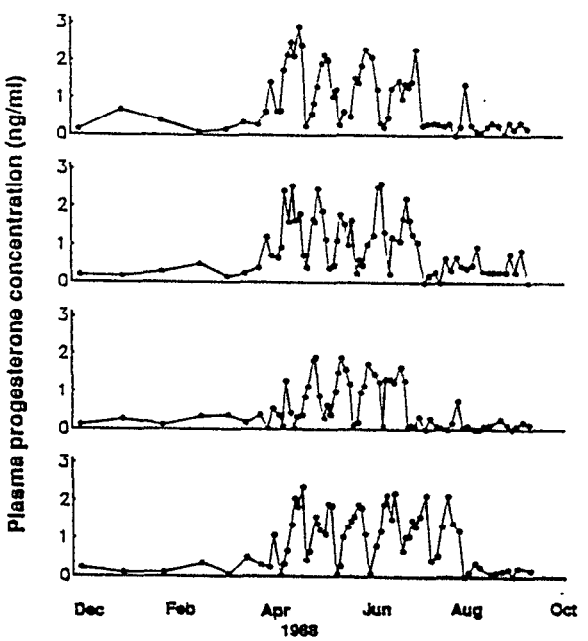
Mean live weight (Fig 1) increased from 77.8 ± 0.8 kg to 86.6 ± 1.0 kg between December 1987 and March 1988 and remained between 86 and 90 kg during autumn and early winter. In early August there was a sharp decrease (6 kg) in mean live weight to about 84 kg followed by a rise in late spring to 91.5 ± 1.5 kg.

FIGURE 1 Mean live weight (kg) of pubertal red deer hinds (n=4) from December 1987 to October 1988. Vertical bars denote s.e.m.



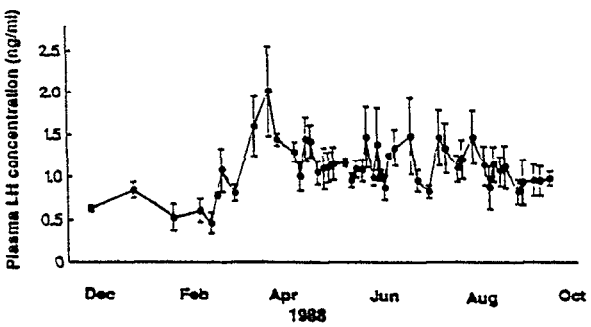
Plasma progesterone concentrations (Fig 2) were low (about 0.5 ng/ml) until early April. In mid April (15 April ± 2.4 d) a transient (<3 d) increase in plasma progesterone concentration (> 1 ng/ml) was recorded in each hind and was followed by the first sustained increase (> 1 ng/ml for 9-13 d) indicating a full luteal cycle on 26 April ± 3.4 d. Individual plasma progesterone profiles showed that 4-6 ovulatory cycles occurred in each hind. Mean ovulatory cycle length (time from increase in progesterone concentration > 1 ng/ml until beginning of the subsequent luteal phase) was 18.6 ± 0.9 d. The period of ovarian cyclicity (time from the beginning of the first luteal phase to the end of the last luteal phase) varied considerably between hinds, ranging from 64 to 107 d (86.5 ± 8.9 d) with each final cycle ending in July or August (21 July ± 7.2 d).

FIGURE 2 Individual plasma progesterone concentrations of pubertal red deer hinds from December 1987 to October 1988.



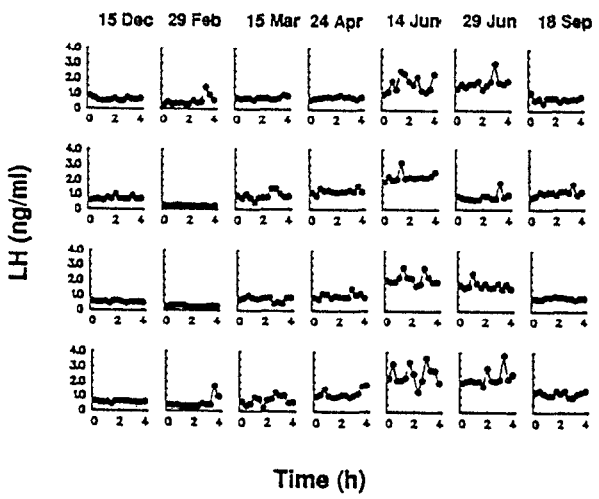
Plasma LH concentrations of the infrequent blood samples varied considerably in individual hinds. Mean plasma LH concentration (Fig 3) was generally low (about 0.5 ng/ml) between December and late February but increased in March to peak at 2 ng/ml in early April. Mean plasma LH concentration varied between 0.9 and 1.5 ng/ml thereafter.

FIGURE 3 Mean plasma LH concentration of pubertal red deer hinds (n=4) from December 1987 to October 1988. Vertical bars denote s.e.m.



During the intensive sampling periods (Fig 4), plasma LH concentrations (Table 1) were significantly greater on 14 and 29 June than in December, February and March with values in April, March and September being intermediate between these extremes. The greatest number of LH pulses was detected on 14 June (2.0 ± 0.7 pulses/4 h), significantly more than in December, February, March or September ($p<0.05$). When the number of pulses during an intensive sampling period was sufficient (i.e. > 1 pulse for > 3 hinds) to permit mean pulse amplitude to be calculated (ie 24 April, 14 and 29 June) it was possible to show that mean pulse amplitude was lower in April than on 29 June ($p<0.05$). Mean plasma LH concentrations during the intensive blood sampling period were positively correlated with LH pulse frequency (Spearman rank correlation coefficient $r=0.66$, $p<0.01$) but there was no relationship between frequency and amplitude of pulses ($r=0.25$, $p>0.05$). Plasma LH concentration always peaked 10 min after the administration of GnRH and the magnitude of the increase did not change with season ($p>0.05$) (Table 1).

FIGURE 4 Individual plasma LH profiles of pubertal red deer hinds intensively sampled on 7 occasions between December 1987 and October 1988.



Most (25/28) of the entire-mated hinds from the same age cohort but not involved in the intensive study calved as 2-year-olds in December (mean calving date, 16 December ± 3.6 d), significantly later than untreated mature hinds on the same property (23 November ± 1.5) ($p<0.05$). Estimated conception dates, based on a gestation length of 233 d (Kelly & Moore, 1977), of these 2-year-old and adult hinds were 27 April and 4 April, respectively.

DISCUSSION

Plasma progesterone profiles indicated that, in the yearling red deer hinds, ovulatory cycles were initiated in late April and terminated between July and August, 3 months later. The duration of the breeding season therefore appears to be shorter in pubertal hinds than in adults, as unmated mature hinds cycle for about 5 months, with ovarian cycles beginning in early April (October in the Northern Hemisphere) and continuing until September (March in the Northern Hemisphere) (Curlewis *et al.*, 1988; Loudon, *et al.*, 1989; Adam *et al.*, 1989; Meikle *et al.*, 1991). One factor contributing to the short duration of the breeding season of pubertal red deer hinds was the later onset of ovarian activity recorded in young hinds, as observed in this study and by Loudon *et al.* (1989). This was reflected in the calving data of 2-year-old hinds of the same age cohort which calved 3 weeks later than the older hinds. The early onset of seasonal anoestrus also shortened the duration of the breeding period in yearling hinds. Observational data from Guinness *et al.* (1971) showed that the mating season of captive yearling red deer hinds in Scotland was only about 3 months long. Oestrous behaviour ceased for most pubertal hinds during December (June, Southern Hemisphere) and, as in the present study, the date of the last cycle varied greatly between hinds (17 November-20 March). Late onset of reproductive activity and early onset of seasonal anoestrum in pubertal animals has also been reported in the ewe lamb (Hafez, 1952; Foster & Ryan, 1981) and fallow deer doe (Asher, 1985). The short breeding period decreases the opportunities for seasonally breeding animals to conceive in their first season. If the onset of breeding activity is delayed due to poor nutrition or to late date of birth the young female may remain anovulatory until the following breeding season (Foster, 1988). This situation is unlikely to arise in well-grown red deer yearlings on New Zealand farms but may explain the impaired reproductive performance of young red deer hinds in the wild (Clutton-Brock *et al.*, 1982).

Hinds in the present study gained weight during summer and early autumn, exceeding the threshold live-weight range for puberty of 65-70 kg (Kelly & Moore, 1977; Hamilton & Blaxter, 1980; Fisher & Fennessy, 1985) throughout the trial period. A large decrease in live weight was experienced in late winter when feed reserves ran short and the feed requirements of other animals on the Deer Unit had higher priority. However, it is unlikely that this loss of live weight influenced the cessation of reproductive activity in the trial as 3 of the 4 hinds had ceased to cycle prior to the onset of the feed shortage.

Puberty in these yearling red deer hinds was characterised by a short-term increase in progesterone concentrations followed by 4-6 full-length ovarian cycles, lasting approximately 19 days each. The progesterone profile of each cycle was typical of the progesterone pattern observed in adult hinds during the breeding season (Adam *et al.*, 1989; Jopson *et al.*, 1990). In lambs the first LH surge, marking the pubertal transition to breeding activity, is also associated with a short term rise in plasma progesterone level which is followed by a second LH surge and a normal luteal-phase increase in circulating progesterone concentration (Foster *et al.*, 1986). Since the plasma LH surge associated with ovulation is of short duration (< 24 h, Duckworth & Barrell, 1991) the relatively long (2 to 3 day) sampling interval used in this study would not reliably detect the preovulatory rise in plasma LH concentration. Minor increases in mean plasma LH concentration were detected at the onset of the breeding season and were similar to the transient peaks of LH which have been detected in plasma samples collected from sheep (I'Anson & Legan, 1988) and pubertal heifers (Gonzales-Padilla *et al.*, 1975) before the onset of the breeding season. Endocrine changes around puberty in red deer hinds appear to be similar to those accompanying the onset of the breeding season in adult hinds (Jopson *et al.*, 1990) and pubertal and adult ewes (Foster, 1988; Goodman, 1988). Progesterone profiles indicate that all hinds in April and June were intensively blood sampled during the luteal phase of the oestrous cycle. All other intensive blood samplings occurred whilst hinds were acyclic. As in the ewe lamb (Foster *et al.*, 1975; Huffman *et al.*, 1987), LH pulse frequency was low (< 1 pulse/ 4 h) in the hinds before puberty and relatively high (about 1-2

TABLE 1. LH secretion in young red deer hinds during intensive blood sampling periods and in response to GnRH challenge between December 1987 and October 1988. Means within columns assigned different superscripts are significantly different ($p < 0.05$)

Date	Mean plasma LH (ng/ml)		Pulse frequency (pulses/4h)		Pulse amplitude (ng/ml)		LH response to GnRH (ng/ml)	
	mean	s.e.m.	mean	s.e.m.	mean	s.e.m.	mean	s.e.m.
15 December 1987	0.63^{ab}	0.04	0.25^{a}	0.25	-	-	2.28^{a}	0.52
29 February 1988	0.38^{a}	0.08	0.50^{ab}	0.29	-	-	3.97^{a}	1.11
15 March 1988	0.78^{abc}	0.04	0.50^{ab}	0.29	-	-	2.60^{a}	0.48
24 April 1988	1.05^{bcd}	0.10	1.25^{bc}	0.48	0.43^{a}	0.02	3.60^{a}	0.58
14 June 1988	1.85^{e}	0.29	2.00^{c}	0.42	0.82^{ab}	0.17	3.92^{a}	0.41
29 June 1988	1.69^{de}	0.30	1.25^{bc}	0.25	1.11^{b}	0.11	3.86^{a}	0.55
18 September 1988	1.27^{cde}	0.30	0.25^{a}	0.25	-	-	4.55^{a}	0.40

pulses/4 h) during the breeding season. The increase in LH pulsatility at the beginning of the breeding period presumably plays a central role in the initiation of ovarian cyclicity in the hind as it does in the pubertal and adult ewe (Foster, 1988; Goodman, 1988).

In contrast to the adult Père David's deer hind (Curlewis *et al.*, 1991) and the lactating red deer hind (Duckworth and Barrell, unpublished), the LH response to exogenous GnRH in young red deer hinds here was not reduced during the anovulatory period which makes it unlikely that the low pulse frequency of LH recorded before puberty was due to failure of the pituitary to respond to endogenous GnRH. Rather the seasonal differences in LH secretion can be explained by seasonal changes in the output of GnRH from the hypothalamus.

These results indicate that, as for other seasonally breeding animals such as fallow deer (Asher, 1985) and sheep (Hafez, 1952; Dyrmundsson, 1973), the breeding season of pubertal red deer hinds is shorter than that of older hinds.

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REFERENCES

Adam, C.L.; Moir, C.E.; Shiach, P. 1989. Melatonin can induce year-round ovarian cyclicity in red deer (*Cervus elaphus*). *Journal of Reproduction and Fertility*, **87**: 401-408.

Asher, G.W. 1985. Oestrous cycle and breeding season of farmed fallow deer, Dama dama. *Journal of Reproduction and Fertility*, **75**: 521-529.

Clutton-Brock,T.H.; Guinness, F.E.; Albon, S.D. 1982. *In* Red deer. Chicago, The University of Chicago Press.

Curlewis J.D.; Loudon, A.S.I.; Milne, J.A.; McNeilly, A.S. 1988. Effect of chronic long acting bromocriptine treatment on live weight, voluntary feed intake, coat growth and breeding season in non- pregnant red deer hinds. *Journal of Endocrinology*, **119**: 413-420.

Curlewis, J.D.; McLeod, B.J.; Loudon, A.S.I. 1991. LH secretion and response to GnRH during seasonal anoestrus of Père David's hinds (*Elaphurus davidianus*). *Journal of Reproduction and Fertility*, **91**: 131-138.

Duckworth, J.A.; Barrell, G.K. 1991. Oestrous behaviour and luteal function in anoestrous red deer hinds treated with a GnRH analogue or oestradiol. *Proceedings of the New Zealand Society of Animal Production*, **51**: 55-61

Dyrmundsson, O.R. 1973. Puberty and early reproductive performance in sheep.1. Ewe lambs. *Animal Breeding Abstracts*, **41**: 273-289.

Fisher, M.W.; Fennessy, P.F. 1985. Reproductive physiology of female red and wapiti deer. *Proceedings of a Deer Course for Veterinarians, Deer Branch Course No. 2, Deer Branch of New Zealand Veterinary Association*, pp. 88-100.

Foster, D.L. 1988. Puberty in the female sheep. *In* The Physiology of Reproduction, Eds E. Knobil & J. Neill *et al.*. pp 1739-1762. Raven Press Ltd, New York.

Foster, D.L.; Karsch, F.J.; Olster, D.H.; Ryan, K.D.; Yellon, S.M. 1986. Determinants of puberty in a seasonal breeder. *Recent Progress in Hormone Research*, **42**: 331-384.

Foster, D.L.; Lemons, J.A.; Jaffe, R.B.; Niswender, R.B. 1975. Sequential pattern of circulating luteinizing hormone and follicle-stimulating hormone in female sheep from early post- natal life through the first estrous cycles. *Endocrinology*, **97**: 985-994.

Foster, D.L.; Ryan, K.D. 1981. Premature seasonal inhibition of tonic LH secretion by oestradiol in the female lamb and its consequences. *Journal of Reproduction and Fertility*, **63**: 289-294.

Foster, D.L.; Yellon, S.M.; Olster, D.H. 1985. Internal and external determinants of the timing of puberty in the female. *Endocrinology*, **75**: 327-344.

Goodman, R.L. 1988. Neuroendocrine control of the ovine estrous cycle. *In* The Physiology of Reproduction, Ed E. Knobil & J. Neill *et al.*. pp 1929-1969. Raven Press Ltd, New York.

Gonzalez-Padilla, E.; Wiltbank, J.N.; Niswender, G.D., 1975. Puberty in beef heifers. I. The interrelationship between pituitary, hypothalamic and ovarian hormones. *Journal of Animal Science*, **40**: 1091-1104.

Guinness, F.E.; Lincoln, G.A.; Short, R.V. 1971. The reproductive cycle of the female red deer, *Cervus elaphus L. Journal of Reproduction and Fertility*, **27**: 427-438.

Hafez, E.S.E. 1952. Studies on the breeding season and reproduction in the ewe. *Journal of Reproduction and Fertility*, **42**: 189-265.

Hamilton, W.J.; Blaxter, K.L. 1980. Reproduction in farmed red deer. I. Hind and stag fertility. *Journal of Agricultural Science*, **95**: 261-275.

Huffman, L.J.; Inskeep, E.K.; Goodman, R.L. 1987. Changes in episodic luteinizing hormone secretion leading to puberty in the lamb. *Biology of Reproduction*, **37**: 755-761.

I'Anson, H.; Legan, S.J. 1988. Does the first LH surge of the breeding season initiate the first full-length cycle in the ewe. *Journal of Reproduction and Fertility*, **82**: 761-767.

Jopson, N.B.; Fisher, M.W.; Suttie, J.M. 1990. Plasma progesterone in cycling and in ovariectomised red deer hinds: the effect of progesterone supplementation and adrenal stimulation. *Animal Reproduction Science*, **23**: 61-73.

Kelly, R.W.; Moore G.H. 1977. Reproductive performance in farmed red deer. *New Zealand Agricultural Science*, **11**: 179-181.

Loudon, A.S.I.; Milne, J.A.; Curlewis, J.D.; McNeilly, A.S. 1989. A comparison of the seasonal hormone changes and patterns of growth, voluntary food intake and reproduction in juvenile and adult red deer (*Cervus elaphus*) and Père David's deer (*Elaphurus davidianus*) hinds. *Journal of Endocrinology*, **122**: 733-745.

Meikle, L.M.; Fisher, M.W.; McLeod, B.J.; Whaanga, W.J.; Johnstone, P.D. 1991. Basal and GnRH-induced secretion in the red deer hind: the effects of ovariectomy and oestradiol treatment. *Proceedings of the 23rd Annual Conference of the Australian Society of Reproductive Biology.* Abstr. 41

Webster, J.R.; Barrell, G.K. 1985. Advancement of reproductive activity, seasonal reduction in prolactin secretion and seasonal pelage changes in pubertal red deer hinds (*Cervus elaphus*) subjected to artificially shortened daily photoperiod or daily melatonin treatments. *Journal of Reproduction and Fertility*. **73**: 255-260.