Role of thyroid hormones in the control of seasonal breeding in red deer

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Abstract

It is assumed that the control of seasonal breeding in female deer follows the model developed from studies in sheep, in which the annual breeding season involves high frequency discharges of GnRH secretion by the hypothalamus which are intermittently inhibited by progesterone secreted from the corpus luteum. In the non-breeding season, the frequency of discharge of GnRH is low because the hypothalamus is sensitive to inhibition by oestrogens from the ovaries and because of other, 'steroid independent', changes in hypothalamic function. As a result oestrous cycles do not occur. Alternation between the two reproductive states can occur spontaneously but is entrained to a twelve-monthly cycle by an action of the increase in daily photoperiod each spring. Studies in sheep and red deer stags have shown that the annual transition to the non-breeding state is dependent on the presence of thyroid hormones and this finding has been extended to red deer hinds by recent work at Lincoln University. In the case of red deer hinds the involvement of thyroid hormones is directed at the steroid independent inhibition of reproduction. It is possible that this finding may lead to extension of the breeding season of commercially farmed deer once a method for temporarily switching off thyroid gland secretion has been developed.

Key words: seasonal breeding, thyroidectomy, red deer, luteinizing hormone, ovariectomy

Introduction

The recent work on reproduction in red deer carried out at Lincoln University has revealed a new avenue for pursuing out-of-season breeding in this species. However, before attempting to describe these studies it will be useful to provide the following update on the control of seasonal breeding.

Breeding seasons

Much of our current understanding of the biology of seasonal breeding in red deer is based on a working model derived from studies on a variety of mammalian species. Obviously this model is subject to constant revision and modification but some of its tenets are well established and it will be useful to set them out here.

Let us be absolutely clear about what the breeding season means to some animals. It is a period when the secretion of hormones, namely LH and FSH from the anterior pituitary gland, is high. That is, these gonadotrophic hormones are secreted in large doses and, especially in the case of

LH, at high pulse frequencies. This situation arises from enhanced secretory activity of the pituitary gland and from the high level of stimulatory activity in the brain which causes the hypothalamus to deliver gonadotrophin-releasing hormone (GnRH) pulses to the pituitary gland at a high frequency. It is convenient to view the hypothalamus as the site of a 'pulse generator', a group of neurons whose function determines GnRH pulse frequency. High frequency GnRH pulses stimulate the pituitary gland and the resulting hypersecretion of LH and FSH stimulates the testes causing increased spermatogenesis and testosterone secretion, the latter of which probably accounts for the well documented effects on behaviour and antler hardening in stags. In females, the extra supply of LH and FSH causes a change in function of the ovaries. Hitherto they have been growing and maturing follicles with resultant follicular death - atresia - but, with the increase in gonadotrophic hormone concentrations in blood, the follicular maturation process culminates in successful ovulation and formation of a corpus luteum. With its formation the corpus luteum secretes progesterone, one of the actions of which is to limit release of GnRH from the hypothalamus. The breeding season is thus characterised by high frequency pulsing of GnRH and its feedback inhibition by progesterone; in short, control of reproduction at this time arises from an interaction between the brain and the ovary.

In contrast, the non-breeding season is characterised by a diminished frequency of GnRH pulses arising from a downturn in stimulatory activity of the brain and also from a heightened sensitivity of the brain to feedback inhibition by steroids - especially oestradiol in females. Again, much of the control emanates from an interaction between the brain and the ovary but this time it involves oestrogens issuing from ovarian follicles. At the moment we have to presume that this is the case in red deer, and the changes in plasma LH concentration described by Miekle and Fisher (1996) for entire and ovariectomized red deer hinds provide strong evidence to support this view.

Control of breeding seasons by daylight

What controls the change between these two reproductive states - breeding and non-breeding? The answer most people give to this questions is - 'daylength'. In a way they are partly correct but we can be far more precise than this. Currently it is believed that some animals - those that display breeding seasonality - can simply oscillate between the two breeding states without the cues of changing daylength. In sheep this has been termed as the 'endogenous circannual rhythm of reproduction' (Karsch *et al.*, 1989) which arises from within the brain. It is called circannual because it has a cycle of 'about' 12 months, i.e. 'circa annual'. Of course, what we see in sheep and other seasonal breeders is a breeding cycle with a length of *exactly* 12 months because the endogenous circannual rhythm has been 'entrained' so that it follows the 12 month cycle of changes in daylight. It is thought that this cueing of the brain is achieved by the daily increase in length of sunlight which occurs each spring (Malpaux *et al.*, 1989). The daylight hours of spring behave rather like a switch which re-sets the endogenous rhythm to the correct 12-months cycle, even though the effect of this switching is not seen externally until six months later - in autumn when breeding activity commences (see Figure 1).

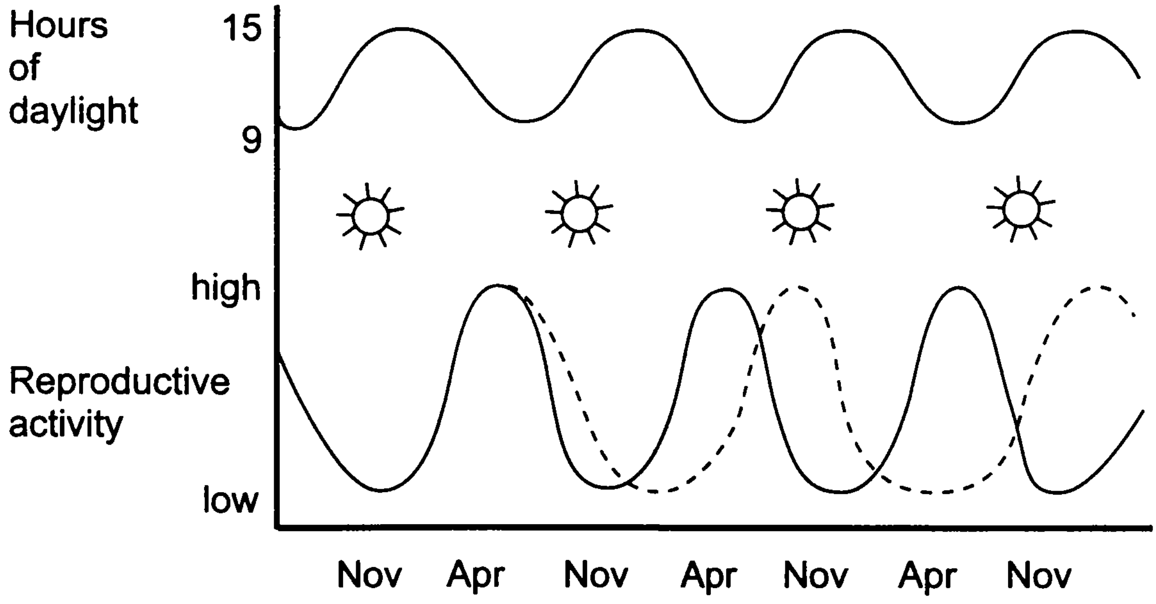

Figure 1. Reproductive activity in a 'short-day' breeding mammal. Note how the lengthening daily photoperiods in spring entrain the breeding season to each autumn (—). In the absence of such daylight cues, animals express their built-in, endogenous, rhythm of reproductive activity (---) so that within a few years their breeding activity may be several months out of phase with the usual annual pattern.

So, even though sheep are often called 'short day' breeders, which is correct in the sense that they do breed in autumn-winter, they are actually most affected in terms of control of reproduction by the long days of spring. This does not negate the fact that reproductive activity can be stimulated directly by short or decreasing daily photoperiods. In fact many people are aware that reproductive activity in sheep and other autumn breeders can be stimulated by subjecting them to artificially shortened photoperiods. However, in spite of this, the correct timing of the annual cycle of reproduction under natural conditions is regulated by the daylight hours of spring.

If the cueing effect of daylight did not occur, seasonal breeding animals would simply express their individual endogenous rhythms of reproduction. Because these are not exactly 12 months, the reproductively active period would eventually occur at any time of the year. For instance an animal with an endogenous rhythm one month different from 12 months would be completely out-of-phase with the norm every six years!

Role of thyroid hormones

In the last decade it has become clear that thyroid hormones have an important role in the control of seasonal breeding (see review by Karsch *et al.,* 1995). Sheep and red deer which have had their thyroid glands surgically removed do not undergo the transition from the breeding to the non-breeding state. This means that animals thyroidectomized during the breeding season remain permanently reproductively active. Obviously, such a finding has potential repercussions for the control of early calving in farmed red deer and this explains why the deer research group at Lincoln University has been carrying out the studies described below.

Insights gained from the thyroidectomized stag

Our first awareness of the role of the thyroid glands in deer reproduction came about from the visit to Lincoln University in 1980 by Tony Care, then Professor of Animal Physiology at the University of Leeds, United Kingdom. He was interested in the possible role of parathyroid gland function in hardening of antlers. To do this parathyroid and thyroid glands were removed surgically from a couple of stags and their antlers were removed later for analysis of bone density (Care *et al*, 1985). After Professor Care returned to the United Kingdom the university was left with two stags which surprised us by surviving in apparent good health for many years and by remaining in a constant rutting state. That is, their antler buttons remained hard, they roared throughout the whole year and their coat moulting lost its usual seasonal pattern.

This unexpected result led us take on a student, Zhendan Shi from China, to carry out a series of studies towards his PhD. Shi made some important discoveries. His work showed that thyroidectomy alone caused red deer stags to remain in the rut state. Their plasma testosterone concentrations did not drop in the spring, nor did their testes diameter fall at this time (see Figure 2).

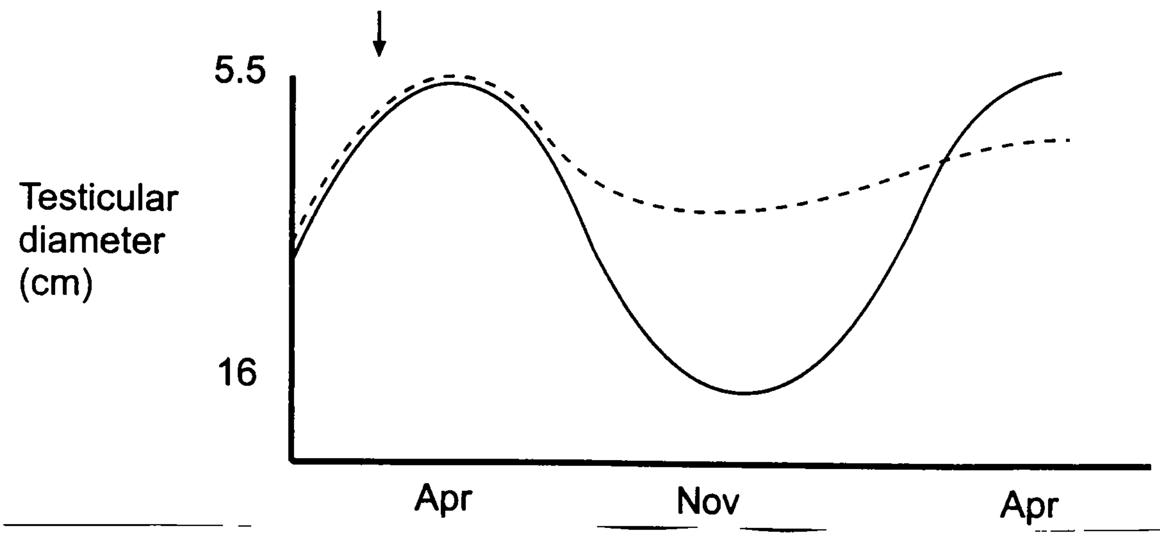

Figure 2. Seasonal pattern of changes in testicular diameter of euthyroid (—) red deer stags and stags thyroidectomized (---) at the time marked with an arrow. (From Shi & Barrell 1992)

They maintained hard antlers and enlarged neck muscles, had delayed coat moults and kept up roaring and generally aggressive behaviour (Shi & Barrell, 1992). Furthermore it was possible to reverse these effects of surgical thyroidectomy by simply injecting thyroid hormones (Shi & Barrell, 1994). This proved that the normal transition to the non-breeding state in spring was dependent on the presence of thyroid hormones.

It is pure coincidence that a similar finding with respect to the breeding season of ewes and involvement of thyroid hormones was made at about the same time in the United Kingdom

(Nicholls *et al.*, 1988) and in Michigan, United States of America (Moenter *et al*, 1991). At this time we suspected that the finding might be generally true for seasonally breeding mammals - the transition from the breeding to the non-breeding state is dependent on the presence of thyroid hormones, albeit in a permissive role (Dahl *et al.*, 1995). So, is this true for red deer hinds?

Thyroid function and the breeding season in red deer hinds

Our studies on the role of thyroid hormones in the control of seasonal breeding in red deer hinds formed the basis of the PhD programme of Greg Anderson. His initial approach to the question was to look at the seasonal pattern of LH secretion in hinds which had been thyroidectomized. Unfortunately the cyclic nature of ovarian function causes changes in LH secretion which make such studies on entire hinds difficult to interpret so Greg ovariectomized the hinds as well. This, of course, removed the source of oestrogens and it is important to maintain blood levels of oestrogens at about the normal level to keep the animals within physiological bounds. Subcutaneous implants which provided an unvarying source of oestradiol were placed in the thyroidectomized/ovariectomized hinds to overcome this problem and LH concentrations in plasma were monitored at intervals throughout the year.

In contrast to what we would have expected from the results of the stag and sheep studies, these thyroidectomized hinds reduced blood LH concentrations in spring just as if they were undergoing the end of breeding activity. This result in thyroidectomized hinds was surprising as it indicated that deer hinds must be different from sheep and stags as far as the role of thyroid hormones is concerned. However, since then Greg Anderson went on to show that the picture changed when the oestradiol implants were removed. In the absence of oestradiol implants thyroidectomized/ovariectomized hinds elevated blood concentrations of LH in spring (see Figure 3), when thyroid intact ovariectomized hinds experienced suppression of LH concentrations. This showed that the red deer hind also requires thyroid hormones for the termination of the breeding season in spring but in this animal the action is aimed at the brain mechanism which controls the 'steroid-independent' component of seasonality - hence no noticeable effect when the oestradiol implants were in place. So, in a sense, the hinds are not so different from stags and sheep after all. It is just that the relative importance of the brain-derived control of seasonality and that arising from negative feedback by steroids differs between these animals. Follow up experiments using ovary-intact hinds showed that thyroidectomy during the breeding season allowed oestrous cycles to continue throughout the following spring (see Figure 4) and that this effect of thyroidectomy could be reversed by treating hinds with thyroxine. So, whether they have an ovary or not, red deer hinds switch off reproductive activity in spring and this can be prevented by thyroidectomy.

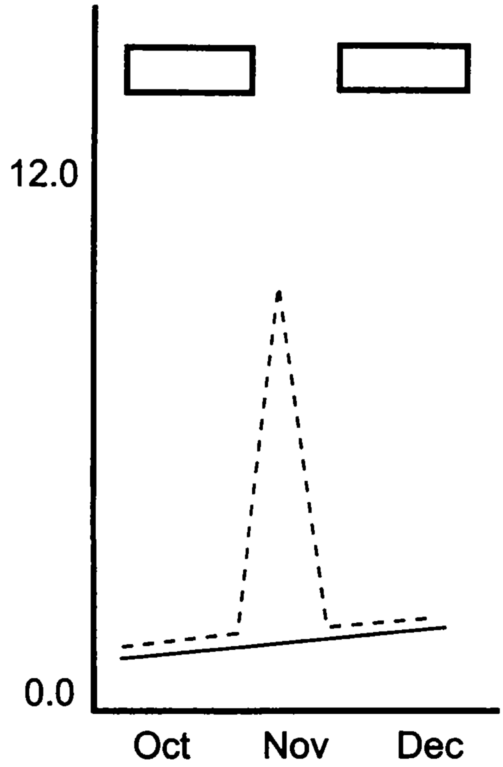

Figure 3. Plasma luteinizing hormone(LH) concentrations (ng/ml) of ovariectomized red deer hinds which were euthyroid (—) or thyroidectomized in May (---). Blocks at the top indicate when s.c. implants containing oestradiol were present. (From Anderson & Barrell, 1998)

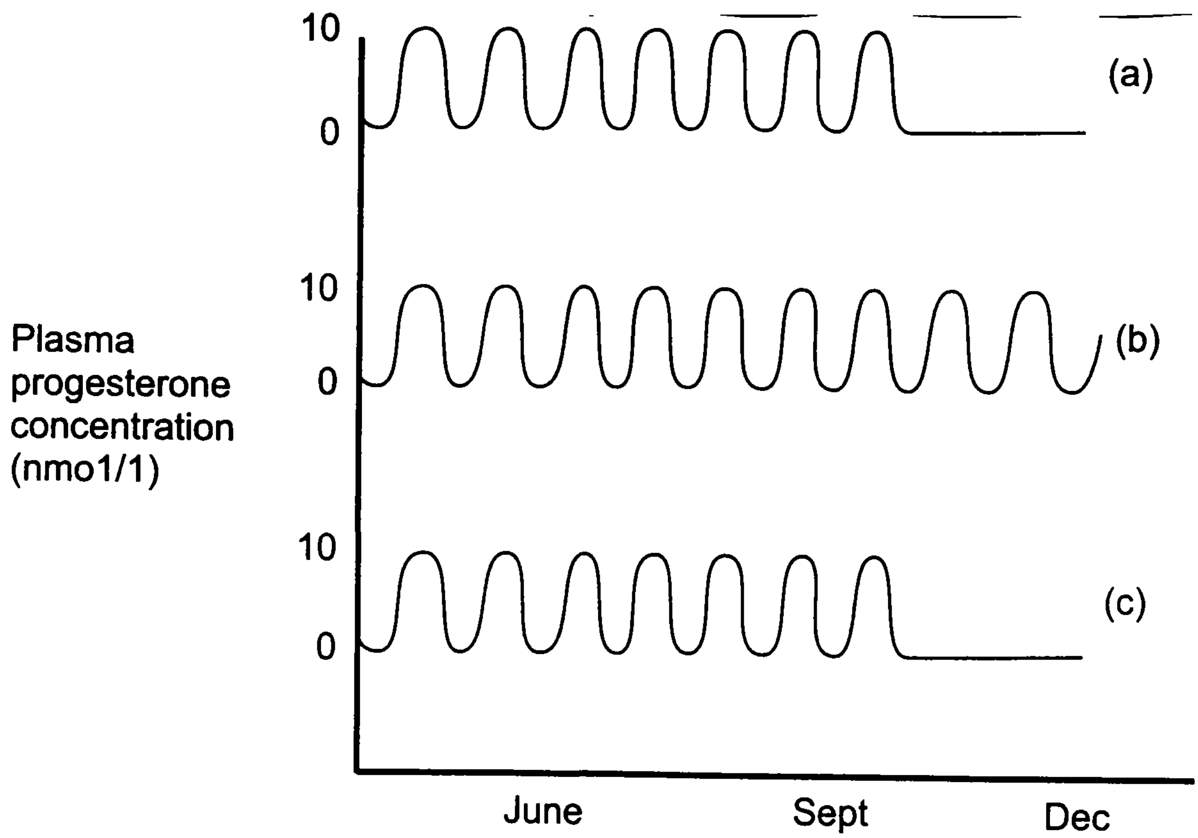

Figure 4. Patterns of plasma progesterone concentration in unmated red deer hinds which were: (a) - euthyroid, (b) - thyroidectomized in May, or (c) - thyroidectomized in May but treated with s.c. implants containing thyroxine. (From Anderson & Barrell 1998)

Greg has gone on to demonstrate that thyroidectomized hinds can be mated in December with some (40%) success in conception and give birth to live calves in August. Interestingly some thyroid-intact hinds also conceived and calved in concert with these hinds so it is possible that the thyroidectomized animals induced breeding activity in the normal hinds by a social facilitation effect.

Prospects for feasible on-farm application

It must be stressed here that reproduction in thyroidectomized animals is not very successful and we do not envisage any on-farm use of such animals. We have used them purely as research tools to help us to understand the biology of the control of seasonal breeding - in particular the role of thyroid hormones in this process. Nevertheless, the results of these studies do pave the way to techniques for advancing the breeding season on deer farms. Recent studies on sheep (Thrun *et al*, 1997) and our work on deer (Anderson & Barrell, 1998) indicate that there is a short period of time - a 'window' - when the reproductive system is sensitive to the switching off effect of thyroid hormones. It may be possible to use a simple injection of a drug to cause temporary suppression of thyroid gland activity during the 'window' period. This could enable otherwise normal hinds to maintain reproductive activity outside the usual breeding season and might be a very effective application of our findings. Currently we are experimenting with techniques to achieve temporary inhibition of thyroid function (Saleh *et al.*, 1998). At the moment we are using sheep for this work simply for convenience. If we achieve a breakthrough we will try it out on deer straight away as we see this as the species for which such technology has an immediate use at the farm level.

Acknowledgements

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