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THE USE OF LIGHT AND MELATONIN TREATMENTS IN THE PREPARATION OF SUFFOLK RAMS FOR OUT-OF-SEASON BREEDING

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INTRODUCTION

Lincoln and Davidson (1977) described the sequence of reproductive responses which occur in Soay rams following photostimulation. Several forms of photostimulation, involving a sequence of long and short days, are effective in modifying seasonal fluctuations in the reproductive parameters of rams (Jackson and Williams, 1973; Shanbacher, 1979). In ewes, photostimulation can initiate oestrous cycles during June/July and thus provide the opportunity for autumn lambing (Williams, 1977). Attempts to dispense with a priming period of 'long days' prior to the abrupt introduction of 'short days' were not successful for out-of-season breeding in Suffolk ewes (Williams and Ward, 1988). Conventional light treatments require controlled environment housing to provide 'long nights' and this has curtailed their widespread use, largely due to the cost of providing specialized housing and to the disruption of grazing management from April to June.

It has been established that melatonin is produced by the pineal gland only during the period of darkness and that it plays a key rôle in the periodicity of Soay rams (Almedia and Lincoln, 1984) and of Suffolk ewes (Arendt, Symons, Land and Pryde, 1983). Recent investigations have shown that melatonin treatment may replace the 'long night' phase of light treatments, and that a high level of fertility may be achieved in adult Suffolk ewes (Williams, 1984, 1985; Williams and Ward, 1988). It greatly simplifies the management of ewes bred out-of-season. Hanif and Williams (1988) have demonstrated that melatonin implants (Regulin®, Regulin Ltd, Australia) are an effective alternative to 'long nights' in the treatment of yearling Suffolk rams for out-of-season breeding. The main objectives of this investigation were the comparison of timed and continuous (implant) administration of melatonin, and the evaluation of the rôle of a priming period of 'long days' preceding melatonin treatments applied to yearling Suffolk rams from mid March.

TABLE 1
Treatments† and period

Group	No.	1 February to 14 March	14 March to 9 September
A	8	18L : 6D	LL + melatonin implant‡
B	8	18L : 6D	LL + melatonin fed§
C	8	LL	LL + melatonin implant
D	8	LL	LL

† LL = local light at 51°43′N; L = hours of light; D = hours of dark.
‡ Melatonin implant = Regulin® (Regulin Ltd, Australia).
§ Melatonin = M – 5250 (Sigma Chemical Company, USA).

MATERIAL AND METHODS

Thirty-two yearling Suffolk rams were allocated to four treatment groups (A, B, C and D) on 1 February 1988. The treatments and their duration are shown in Table 1.

The rams were housed throughout the investigation and penned in subgroups of four rams. They were maintained on dried grass cubes, proprietary compound foodstuffs and mineral supplements according to recommended rates. Melatonin implants were inserted subcutaneously near the base of the ear at 5-week intervals.

Group B were group fed 100 g pelleted food per ram at 16.00 h daily. This provided approximately 3 mg melatonin per ram. Testes measurements were made fortnightly. Sexual behaviour tests were carried out fortnightly; each ram was exposed to two treated ovariectomized ewes displaying oestrus for 10 min, in a boarded test pen 100 m^2. Quality assessments were carried out on semen samples, collected by artificial vagina, fortnightly. Semen collections and behavioural tests were undertaken during alternate weeks throughout the investigation.

RESULTS

Live weight

The main live weight of the four groups at three stages of the investigation are shown in Table 2 and show that there were no significant differences.

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Testis diameter

The mean testis diameter of each group, from the stage when significant differences were first recorded, is presented in Table 3. There were no significant differences between groups A and B at any stage of the investigation. In groups A and B testis diameter increased until early July. Both groups were significantly higher than group D on nine consecutive occasions from 5 April to 26 July. Testis regression commenced in mid July in groups A and B and continued for the remainder of the investigation. In contrast, testis diameter in group D increased from mid June and was maintained for the remaining period; it was significantly higher than groups A and B from 26 August onwards. In group C testis diameter slowly increased until the end of May, but remained significantly lower than groups A and B throughout May. In group C, testis regression occurred from mid June to mid July and was followed by another period of growth.

TABLE 2
Live weight (kg)

	Date					
	27 January		14 June		9 September	
Group	Mean	s.e.	Mean	s.e.	Mean	s.e.
A	56·4	2·56	71·0	2·36	81·0	2·42
B	56·1	2·58	70·4	2·34	82·0	2·08
C	57·0	3·84	69·0	3·16	81·3	3·40
D	56·1	2·47	71·3	1·39	80·5	1·41

Sexual behaviour

The mean number of services achieved per test period from the stage when significant differences occurred is shown in Table 4. Groups A and B achieved higher numbers of services than group D on 11 and 10 consecutive occasions respectively, and were also higher than group C on nine occasions. The performance of group D improved as the breeding season approached.

Semen volume

Table 5 shows the period during which significant differences were recorded in semen volume. Groups A and B did not differ significantly at any stage. Group A produced more semen than groups C and D on six and four occasions respectively; group B produced more than these two groups on six and two occasions. The semen volume of group D increased during the approach to the breeding season.

Sperm concentration

Significant differences between groups in sperm concentration were recorded between 20 April and 6 September (Table 6). Groups A and B did not differ at any stage; both were higher than control group D on eight consecutive occasions and were higher than group C on six occasions. Group C were higher than group D on two occasions during mid April and early May but this was not maintained.

DISCUSSION

The gradual increase in live weight, maintained from the yearling to the 19 month stage, conformed to the expected growth and development for that period. All

TABLE 3
Testis diameter (mm)†

	Group A		Group B		Group C		Group D	
Date	Mean	s.e.	Mean	s.e.	Mean	s.e.	Mean	s.e.
05.4.88	$53{\cdot}50^{c}$	1·38	$52{\cdot}37^{b}$	1·68	$51{\cdot}50^{a}$	1·38	$47{\cdot}00^{abc}$	0·82
19.4.88	$56{\cdot}0^{g}$	1·18	$56{\cdot}0^{h}$	1·40	$53{\cdot}25^{d}$	1·45	$46{\cdot}38^{ghd}$	0·58
03.5.88	$59{\cdot}63^{fb}$	1·54	$59{\cdot}0^{ga}$	1·12	$54{\cdot}75^{abh}$	1·26	$45{\cdot}88^{fgh}$	0·84
17.5.88	$60{\cdot}25^{fb}$	1·60	$62{\cdot}50^{gc}$	1·35	$56{\cdot}13^{bch}$	1·25	$47{\cdot}75^{fgh}$	0·68
31.5.88	$61{\cdot}13^{fb}$	1·58	$63{\cdot}25^{gc}$	1·39	$56{\cdot}50^{bch}$	1·30	$45{\cdot}38^{fgh}$	0·55
14.6.88	$62{\cdot}0^{fc}$	1·56	$63{\cdot}50^{g}$	1·24	$56{\cdot}0^{gc}$	1·07	$45{\cdot}0^{fg}$	0·66
01.7.88	$63{\cdot}50^{g}$	1·24	$65{\cdot}01^{h}$	1·10	$54{\cdot}50^{gh}$	1·38	$47{\cdot}0^{gh}$	0·66
14.7.88	$62{\cdot}0^{fc}$	1·56	$63{\cdot}0^{gh}$	1·10	$53{\cdot}0^{hc}$	1·63	$51{\cdot}0^{fg}$	0·85
26·7·88	$60{\cdot}0^{bc}$	1·56	$61{\cdot}0^{df}$	1·10	$54{\cdot}25^{bd}$	1·66	$55{\cdot}0^{cf}$	0·73
09.8.88	58·50	1·30	59·00	1·10	56·0	1·70	57·00	0·73
26.8.88	$50{\cdot}0^{b}$	1·35	$55{\cdot}0^{c}$	1·07	58·0	1·44	$60{\cdot}0^{bc}$	0·82
09.9.88	$54{\cdot}0^{fb}$	1·35	$53{\cdot}0^{gc}$	1·07	$59{\cdot}0^{bc}$	1·35	$62{\cdot}0^{fg}$	0·82

† Values with similar superscripts within lines are significantly different: a, b = $P < 0{\cdot}05$; c, d = $P < 0{\cdot}01$; f, g, h = $P < 0{\cdot}001$.

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TABLE 4
Number of services per test period†

	Group A		Group B		Group C		Group D	
Date	Mean	s.e.	Mean	s.e.	Mean	s.e.	Mean	s.e.
13.4.88	$2{\cdot}71^{c}$	0·18	$2{\cdot}57^{d}$	0·20	2·37	0·18	$2{\cdot}0^{ac}$	0·0
27.4.88	$2{\cdot}86^{c}$	0·26	$2{\cdot}67^{a}$	0·21	2·38	0·18	$1{\cdot}88^{ac}$	0·23
11.5.88	$2{\cdot}83^{fb}$	0·17	$2{\cdot}83^{ag}$	0·17	$2{\cdot}25^{ab}$	0·16	$1{\cdot}88^{fg}$	0·13
25.5.88	$2{\cdot}86^{ac}$	0·26	$2{\cdot}67^{bd}$	0·21	$2{\cdot}0^{ab}$	0·19	$1{\cdot}50^{cd}$	0·27
08.6.88	$2{\cdot}86^{ce}$	0·26	$2{\cdot}67^{bd}$	0·21	$1{\cdot}88^{eb}$	0·13	$1{\cdot}63^{cd}$	0·18
22.6.88	$3{\cdot}14^{gh}$	0·26	$2{\cdot}86^{ab}$	0·34	$1{\cdot}75^{gb}$	0·16	$1{\cdot}71^{ha}$	0·18
06.7.88	$3{\cdot}29^{ac}$	0·18	$2{\cdot}86^{bd}$	0·14	$2{\cdot}0^{ab}$	0·38	$2{\cdot}0^{cd}$	0·27
20.7.88	$3{\cdot}29^{cd}$	0·18	$3{\cdot}0^{ab}$	0·26	$2{\cdot}25^{bd}$	0·16	$2{\cdot}0^{ac}$	0·19
03.8.88	$3{\cdot}57^{cn}$	0·20	$3{\cdot}25^{de}$	0·16	$2{\cdot}13^{en}$	0·30	$2{\cdot}13^{cd}$	0·30
17.8.88	$4{\cdot}0^{gh}$	0·22	$3{\cdot}50^{cd}$	0·27	$1{\cdot}75^{hc}$	0·37	$2{\cdot}29^{gd}$	0·29
31·8.88	$3{\cdot}50^{gd}$	0·19	$3{\cdot}0^{h}$	0·19	$1{\cdot}50^{ghc}$	0·27	$2{\cdot}57^{cd}$	0·20

† Values with similar superscripts within lines are significantly different: a, b = $P < 0{\cdot}05$; c, d, e, n = $P < 0{\cdot}01$; f, g, h = $P < 0{\cdot}001$.

TABLE 5
Semen volume (ml)†

	Group A		Group B		Group C		Group D	
Date	Mean	s.e.	Mean	s.e.	Mean	s.e.	Mean	s.e.
01.6.88	$1{\cdot}55^{ab}$	0·18	$1{\cdot}28^{cf}$	0·12	$0{\cdot}66^{bc}$	0·11	$0{\cdot}48^{af}$	0·08
15.6.88	$1{\cdot}28^{ac}$	0·21	$1{\cdot}10^{bd}$	0·15	$0{\cdot}50^{ab}$	0·03	$0{\cdot}45^{cd}$	0·04
12.7.88	$1{\cdot}20^{ac}$	0·12	$1{\cdot}08^{d}$	0·20	$0{\cdot}60^{cd}$	0·11	$0{\cdot}75^{a}$	0·15
26.7.88	$1{\cdot}13^{a}$	0·13	$1{\cdot}11^{b}$	0·21	$0{\cdot}63^{ab}$	0·15	0·80	0·12
23.8.88	$1{\cdot}20^{ao}$	0·12	$1{\cdot}15^{b}$	0·15	$0{\cdot}63^{ab}$	0·16	$0{\cdot}83^{o}$	0·09
06.9.88	$1{\cdot}31^{c}$	0·19	$1{\cdot}25^{b}$	0·22	$0{\cdot}61^{abc}$	0·07	$1{\cdot}20^{a}$	0·16

† Values with similar superscripts within lines are significantly different: a, b, o = $P < 0{\cdot}05$; c, d = $P < 0{\cdot}01$; f = $P < 0{\cdot}001$.

TABLE 6
Sperm concentration ($\times 10^{9}$/ml)†

	Group A		Group B		Group C		Group D	
Date	Mean	s.e.	Mean	s.e.	Mean	s.e.	Mean	s.e.
20.4.88	$4{\cdot}12^{a}$	0·26	$4{\cdot}0^{b}$	0·17	$4{\cdot}40^{o}$	0·12	$3{\cdot}42^{abo}$	0·04
04.5.88	$4{\cdot}60^{c}$	0·27	$4{\cdot}68^{d}$	0·34	$4{\cdot}60^{e}$	0·37	$3{\cdot}35^{cde}$	0·04
18.5.88	$4{\cdot}80^{f}$	0·13	$4{\cdot}90^{g}$	0·20	4·18	0·30	$3{\cdot}40^{fg}$	0·04
01.6.88	$4{\cdot}87^{ac}$	0·35	$4{\cdot}91^{gh}$	0·18	$3{\cdot}70^{ag}$	0·18	$3{\cdot}45^{ch}$	0·04
15.6.88	$5{\cdot}13^{ac}$	0·38	$5{\cdot}08^{bo}$	0·47	$3{\cdot}60^{ao}$	0·07	$3{\cdot}50^{bc}$	0·06
12.7.88	$4{\cdot}90^{ce}$	0·26	$4{\cdot}95^{dn}$	0·21	$3{\cdot}53^{cd}$	0·38	$3{\cdot}60^{en}$	0·26
26.7.88	$4{\cdot}86^{ac}$	0·21	$4{\cdot}72^{bd}$	0·26	$3{\cdot}35^{cd}$	0·27	$3{\cdot}67^{ab}$	0·34
23.8.88	$4{\cdot}56^{ce}$	0·15	$4{\cdot}42^{dn}$	0·18	$3{\cdot}54^{cd}$	0·34	$3{\cdot}70^{en}$	0·21
06.9.88	$4{\cdot}32^{c}$	0·15	$4{\cdot}20^{d}$	0·19	$2{\cdot}93^{acd}$	0·27	$4{\cdot}0^{a}$	0·19

† Values with similar superscripts within lines are significantly different: a, b, o = $P < 0{\cdot}05$; c, d, e, n = $P < 0{\cdot}01$; f, g, h = $P < 0{\cdot}001$.

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rams remained in good health throughout the investigation.

It is evident from the data on testis size, sexual behaviour and semen quality, that treatments A and B resulted in a significant advancement in reproductive performance. Timed and continuous administration of melatonin preceded by a priming period of long days, were equally effective in this respect. The advancement achieved in both groups was comparable with that recorded in yearling Suffolk rams in response to photostimulation (18L : 6D; 9L : 15D) and to 'long days) (18L : 6D) followed by melatonin implants (Hanif and Williams, 1988). The uniform responses observed as a result of these treatments reflect the subtle changes which occur at the hypothalamic level and demonstrates the efficacy of melatonin treatments for the preparation of rams for out-of-season breeding.

A full assessment of fertility was outside the scope of this investigation but the criteria of assessment included those which are commonly used to assess rams in artificial insemination studies (Evans and Maxwell, 1986). The advancement of high reproductive performance in rams is comparable with that achieved in Suffolk ewes on similar forms of treatment (Williams, 1985; Williams and Ward, 1988). In terms of reproductive management, the availability of both cyclic ewes and high performance rams during June/July is advantageous for it allows the application of procedures normally practised in autumn tupping flocks. Since winter housing is now prevalent in many areas the 'long days' associated with treatments A and B can be readily applied; the subsequent use of melatonin implants requires no change in conventional management after the ewes are turned out to grass.

The failure of treatments A and B to sustain high reproductive performance beyond early July is in keeping with the observations of Almedia and Lincoln (1984) who showed that rams become refractory after 16 to 20 weeks of exposure to a specific light regime. It is clear from these data that this phenomenon also applies to melatonin treatments. The data indicate that rams treated in this way may not be at peak performance during the following autumn.

There are situations where it may be difficult to apply the priming period of long days. The data for group C indicate that the abrupt application of melatonin in mid March resulted in a weak response in terms of all the criteria of assessment. This suggests that the normal change in the light : dark ratio from the winter solstice to mid March is inadequate priming for treatment C and that the type of treatment is inappropriate in mid March. The same form of treatment applied from early April to adult and yearling Suffolk ewes resulted in low fertility (Williams and Ward, 1988). Further investigation is required on the use of melatonin treatments initiated between the spring equinox and the summer solstice.

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