Aust. J. Biol. Sci., 1985, **38**, 405–10

Effect on Wool Growth of Thyroxine Replacement in Thyroidectomized Merino Rams

S. Maddocks, Y. Chandrasekhar and B. P. Setchell

Department of Animal Sciences, Waite Agricultural Research Institute, Glen Osmond, S.A. 5064.

Abstract

The effect on wool growth of thyroidectomy with subsequent thyroxine replacement at subnormal and supranormal levels has been investigated in Merino rams fed a restricted basal diet. Thyroidectomy without thyroxine replacement caused a greater than 60% reduction in wool growth. Provision of 30% of normal plasma thyroxine concentrations was sufficient to return wool growth to normal. Similarly, complete replacement of plasma thyroxine levels gave normal wool growth while increasing thyroxine concentrations to three times normal increased wool growth to levels slightly above normal. These results provide a possible explanation for the inability of other workers to correlate seasonal thyroxine patterns and annual wool growth variations.

Introduction

It has long been recognized that hormones influence wool growth (Ferguson *et al.* 1965; Wallace 1979). However, there is little understanding of how such influences are produced by various hormones. Attempts by other workers to relate seasonal thyroxine levels to annual variations in wool growth indicated a need to define more clearly the relationship between endogenous thyroxine levels and wool growth.

Thyroid hormones have received much attention since early studies demonstrated that wool growth all but ceased when sheep were thyroidectomized. It is now widely recognized that the presence of thyroid hormones is necessary for normal wool growth and normal follicle development (Ferguson *et al.* 1956). However, the mode of action of thyroid hormones on wool growth is poorly understood. Godfrey and Tribe (1959) demonstrated that thyroxine (T_4) increased the wool growth of animals fed a restricted diet by increasing staple length with no change in fibre diameter. Ferguson (1958) found that wool growth variations in untreated sheep were greater than the increases resulting from the administration of T_4. As Wallace (1979) mentions, there is also no relationship in untreated sheep between wool growth and metabolic rate.

Recently interest has focused on the similarity between the annual wool growth cycle and the annual cyclic variations in plasma T_4 levels. However, while Reklewska (1971), Wallace (1979) and Ryder (1979) have been unable to correlate these two cycles, their apparent relationship is still being viewed by some as important. While elevated plasma T_4 concentrations will stimulate wool growth (Labban 1957; Ferguson 1958) there is no evidence that endogenous variations in T_4 levels affect the wool growth of grazing sheep. Wallace (1979) indicates that an exception may be in those areas where iodine deficiency occurs or where goitrogens are present, but that even in these situations animals tend to maintain normal or elevated plasma triiodothyronine (T_3) concentrations.

0004-9417/85/040405$02.00

In an attempt to define more clearly the relationship between endogenous T_4 levels and wool growth, we have investigated the effect of thyroidectomy with subsequent T_4 replacement therapy at subnormal, normal and supranormal levels, on wool growth in Merino rams fed a restricted basal diet.

Materials and Methods

Twenty Merino rams, 5 years of age and weighing 59 ± 4 (s.e.m.) kg were housed indoors in individual pens from late March to early June. Natural lighting was maintained throughout the period of experimentation with an average light : dark regime of 10 h : 14 h.

Animals were randomly allocated to five treatment groups of four animals per group. One group was used as a control group and underwent sham operations. The remaining animals were thyroidectomized. Of these animals, one group along with the control group received daily subcutaneous injections of alkaline saline [1·0 ml of 0·9% (w/v) NaCl per animal per day]. The other groups received subcutaneous injections of L-thyroxine (Sigma Chemical Co., St Louis, U.S.A.) commencing at the time of thyroidectomy on a daily basis calculated at either 30, 100 or 300% of the mean thyroxine secretion rate of the group determined prior to thyroidectomy. Actual levels administered were 0·033 mg (30%), 0·15 mg (100%) or 0·5 mg (300%) per animal per day.

Water was available *ad libitum*. All animals received the daily ration of 500 g lucerne chaff and 800 g sheep pellets (Milling Industries, Adelaide). The group receiving the 300% T_4 replacement was further supplemented with 300 g of pellets from the second week of the experiment to minimise weight loss. All animals consumed all their ration each day.

Wool Growth Measurements

Animals were tattooed with a 10 by 12 cm area on the midside of their right flank at the time of thyroidectomy. The wool growth within this area was harvested by clipping with fine animal clippers (Oster, No. 40). The patch was clipped 3 weeks after thyroidectomy and commencement of T_4 replacement, and new wool growth on this area was then harvested a further 3 weeks later and weighed to determine greasy weight. At this time measurements of the tattooed area were made with calipers to account for any variations due to enlargement of the tattooed area with time. Clean weights of the oven-dry wool were measured after successive washes in light petroleum (Shell X4) and water (Ferguson *et al.* 1965).

Each time the tattooed patch was clipped, a line of wool over the right shoulder of each animal was banded at skin level with the Durafur Black solution as described by Williams and Chapman (1966). After the final banding these dyebands were left to grow away from skin level before being harvested to allow measurement of staple length growth between dyebands.

Animals were weighed and bled once per week throughout the course of the experiment at approximately the same time of day on each occasion, just before the morning feeding.

Thyroxine Assay

Total plasma T_4 levels were monitored by radioimmunoassay (Burke and Shakespear 1976). Iodinated thyroxine (L-[3′,5′-^{125}I] thyroxine) was obtained from Amersham Australia. L-Thyroxine (Sigma Chemical Co., St Louis, U.S.A.) was used to prepare assay standards from 0 to 240 nM. Assay performance was monitored with the Thyroid Quality Control kit of the Radiochemical Centre (Amersham, England). Interassay and intra-assay variations were approximately 7 and 8% respectively.

Results

Thyroxine Levels

The plasma T_4 levels for each group are shown in Fig. 1 and demonstrate satisfactory achievement of desired plasma levels for each group.

Body Weight

Animals in the control group and the thyroidectomized animals with 100% T_4 replacement showed small increases in weight over the experimental period

(1 ± 0·71 kg and 1 ± 0·58 kg respectively). Animals receiving 300% T_4 replacement gained 0·5 ± 1·4 kg while those receiving only 30% T_4 marginally lost weight (−0·33 ± 0·88 kg) in the 6-week period. None of these changes were significant when compared with the control group. However, the thyroidectomized animals receiving no T_4 supplement gained 4 ± 1·78 kg in the 6 weeks which is significantly different from the control group ($P < 0·01$).

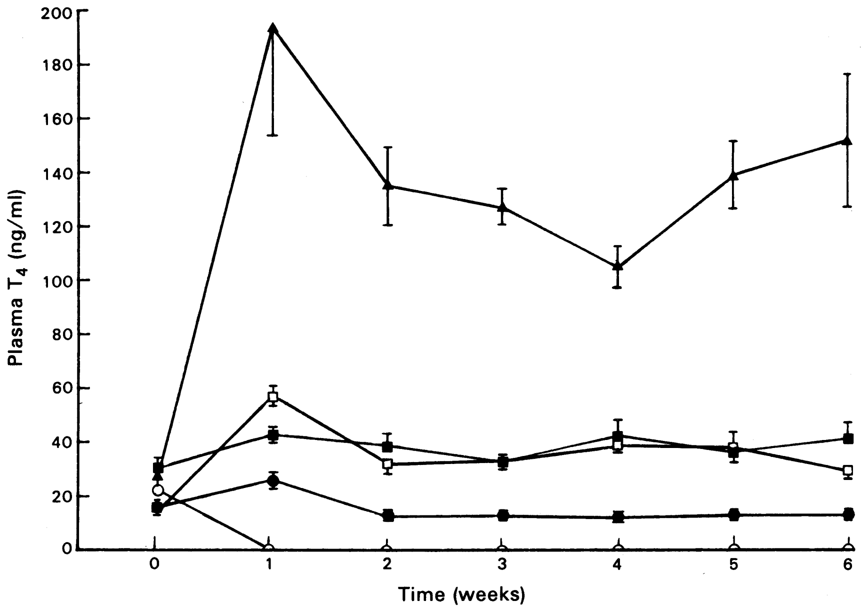

Fig. 1. Plasma thyroxine levels in animals undergoing sham-operations (■), thyroidectomy (○) and thyroidectomy with 30% (●) 100% (□) or 300% (▲) replacement thyroxine. Values are shown as mean ± standard error.

Wool Growth

While the thyroidectomized group receiving no T_4 supplement had a significantly lower ($P < 0·01$) wool production rate over the 3-week period of measurement (dyebands and patch weights), the other treatment groups were not significantly different from the control group, as shown in the following tabulation:

	Control	Thyroid-ectomized	Supplemented 30%	Supplemented 100%	Supplemented 300%
Daily wool production (mg/cm²):	0·58±0·10	0·19±0·10**	0·51±0·14	0·58±0·04	0·65±0·08
Distance between dyebands (mm):	5·0±0·5	0·63±0·06**	5·0±0·5	5·4±0·4	6·3±0·3*

*$P < 0·05$; **$P < 0·01$.

The ratio of weights of clean to greasy wool was not affected by any of the treatments. Staple length growth between dyebands in the thyroidectomized animals receiving 300% T_4 supplement was significantly higher ($P < 0·05$) than the control group. However, the patch weights for the same treatment group were not significantly different from the control group although there was a suggestion of increased growth.

Discussion

In the studies reported in 1958 and 1965 by Ferguson and co-workers, L-thyroxine was administered to thyroidectomized ewes and wool growth was monitored. However, these animals were fed *ad libitum* and received thyroxine doses equivalent to, or greater than, the normal thyroxine secretion rate which was found to be in the region of 0·125 mg per day under the experimental conditions used.

The study reported in this paper is different from these early studies in that we used thyroidectomized rams maintained on a restricted basal diet. The present experiment also appears to be the first in which a much lower than normal amount of T_4 (30%) was administered to one group of animals. The normal thyroxine secretion rate for these rams was found to be in the region of 0·15 mg per day which is comparable to that level determined by Ferguson's group if differences in body weight are taken into account. Although the mean values for plasma T_4 concentrations prior to thyroidectomy are possibly lower than might be expected, individual values ranged from 15 to 50 ng/ml and were consistent for each animal. Assay performance as monitored by quality control samples suggests that the indicated values are real. Breed or age differences may have had an influence on these values. The fact that these rams were on a restricted basal diet, rather than *ad libitum* feeding may also have influenced these initial levels, although weight changes over the experimental period do not reflect an undernourished group of animals.

The provision of thyroxine at levels similar to, or greater than, the normal thyroxine secretion rate has produced results similar to those of Ferguson (1958) and Ferguson *et al.* (1965) but the most significant result of this study is that 30% of normal plasma T_4 levels is sufficient to maintain wool growth at a normal rate. This supports the idea that thyroxine plays a facilitatory role rather than a regulatory role in wool growth. It also provides an understanding of why many workers have been unable to correlate the cyclic T_4 levels in sheep with annual wool growth variations. Ryder (1979) presents data for grazing Soay rams showing an annual T_4 cycle with levels fluctuating between 30 and 80 ng/ml. The lowest point of this cycle is greater than 30% of the highest point of this cycle. This would imply that under normal environmental conditions T_4 levels are rarely low enough to prevent the facilitatory role of T_4 for wool growth. The exception may be the situation described by Wallace (1979) where iodine deficiency or goitrogens markedly impair thyroid function. Lambourne (1964) found that thyroxine implants increased the rate of wool growth in active follicles, while Ryder (1973) found that such treatment failed to restore wool growth in inactive follicles. Unlike Soay sheep, the Merino has little cyclicity in wool growth and wool grows virtually continuously. The results presented in this paper suggest that T_4 is essential to the maintenance of activity in an already active follicle (albeit in small quantities).

The suggestion must be made that thyroxine might be acting as a simple trigger of some cellular event such as that proposed by Turakulov *et al.* (1975) involving mitochondrial protein synthesis, and that the presence of low concentrations of T_4 may be sufficient to allow this process to proceed normally. Alternatively, thyroxine may have a role in affecting nutrient partition. As Black and Reis (1979) indicate in their discussion on nutrient partition between wool growth and other body functions, the rate of wool growth may be affected by hormones which alter the affinity of substrates in various tissues. In the absence of T_4, as produced by the total thyroidectomy of rams in this study, this process becomes severely impaired, and wool growth may all but cease. The possibility that some other factor might be involved in initiating wool growth in previously inactive follicles has not been examined in this study, although the results presented do not preclude this.

The rams of this study were fed a restricted basal diet once per day which may have contributed to the marginal weight loss incurred by the 30% T_4 supplemented group, which nevertheless continued to produce normal amounts of wool. Under the *ad libitum* conditions of grazing, however, these losses might be compensated by increased food intake. Ferguson (1951, 1958) showed that the reduction in wool growth caused by thyroidectomy was not secondary to a depression of food intake. When he fed sheep *ad libitum* and provided thyroxine at doses in excess of 0·5 mg per day the increase in wool growth was accompanied by an increase in feed intake.

While we are still unable to suggest a definitive mode of action for thyroxine on wool growth, the data presented here suggests that thyroxine levels in normal grazing animals are unlikely to ever be of major importance in affecting wool growth since the low levels needed for reduced wool growth are not likely to occur normally in the grazing animal.

Acknowledgments

We are indebted to Mr S. Sowerbutts for his technical assistance, and to Dr C. W. Burke of Oxford University who provided Dr. B. Good with the T_4 antisera. We also wish to acknowledge the assistance and helpful discussions of Dr P. Hynd. Thanks must also go to Mrs S. Suter and Mrs V. Maddocks for their expert typing of the manuscript and to Mr B. Palk for photography. S. Maddocks is supported by an Australian Wool Corporation Post-graduate Scholarship, and Y. Chandrasekhar is supported by a University of Adelaide Research Grant Scholarship.

References

Black, J. L., and Reis, P. J. (1979). Speculation on the control of nutrient partition between wool growth and other body functions. In 'Physiological and Environmental Limitations to Wool Growth'. (Eds J. L. Black and P. J. Reis.) pp. 269–93. (U.N.E. Publication Unit: Armidale.)

Burke, C. W., and Shakespear, R. A. (1976). Triiodothyronine and thyroxine in urine. II. Renal handling and effect of urinary protein. *J. Clin. Endocrinol. Metab.* **42**, 504–13.

Ferguson, K. A. (1951). Ph.D. Thesis, University of Cambridge, England.

Ferguson, K. A. (1958). The influence of thyroxine on wool growth. *Proc. N.Z. Soc. Anim. Prod.* **18**, 128–40.

Ferguson, K. A., Schinckel, P. G., Carter, H. B., and Clarke, W. H. (1956). The influence of the thyroid on wool follicle development in the lamb. *Aust. J. Biol. Sci.* **9**, 575–85.

Ferguson, K. A., Wallace, A. L. C., and Lindner, H. R. (1965). Hormonal regulation of wool growth. In 'Biology of the Skin and Hair Growth'. (Eds A. G. Lyne and B. F. Short.) pp. 655–77. (Angus and Robertson: Australia.)

Godfrey, N. W., and Tribe, D. E. (1959). The effect of thyroxine implantation on wool growth. *J. Agric. Sci.* **53**, 369–73.

Labban, F. M. (1957). The effects of L-thyroxine on sheep and wool production. *J. Agric. Sci.* **49**, 26–50.

Lambourne, L. J. (1964). Stimulation of wool growth by thyroxine implantation. *Aust. J. Agric. Res.* **15**, 657–97.

Reklewska, B. (1975). Wool growth rate in relation to plasma catecholamine and serum total thyroxine concentrations in growing lambs. *Acta Physiol. Pol.* **26**, 461–70.

Ryder, M. L. (1973). A note on the failure of thyroxine to restore wool growth to inactive follicles. *Anim. Prod.* **16**, 319–21.

Ryder, M. L. (1979). Thyroxine and wool follicle activity. *Anim. Prod.* **28**, 109–14.

Turakulov, Y. K., *et al.* (1975). 'Thyroid Hormones. Biosynthesis, Physiological Effects and Mechanisms of Action.' (Consultants Bureau: New York.)

Wallace, A. L. C. (1979). The effect of hormones on wool growth. In 'Physiological and Environmental Limitations to Wool Growth'. (Eds J. L. Black and P. J. Reis.) pp. 257–68. (U.N.E. Publications Unit: Armidale.)

Williams, O. B., and Chapman, R. E. (1966). Additional information on the dye-banding technique of wool growth measurement. *J. Aust. Inst. Agric. Sci.* **32**, 298–300.

Manuscript received 17 May 1985, accepted 23 August 1985