

## Variations in Plasma Thyroxine Concentrations throughout One Year in Penned Sheep on a Uniform Feed Intake

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### Abstract

Plasma total thyroxine ( $T_4$ ) concentrations were measured by radioimmunoassay throughout one year in 10 sheep maintained indoors under natural light and consuming a constant amount of feed. Plasma  $T_4$  concentrations varied throughout the year, the highest and lowest values occurring in the spring and autumn months respectively. The mean of all observations was  $5.4 \pm 0.1$   $\mu\text{g/dl}$ . The changes observed did not appear to correlate with changes in ambient temperature, day length or with reported changes in wool growth.

### Introduction

A limited number of measurements made at different times of the year in small numbers of grazing sheep suggest that there are seasonal variations in plasma thyroxine ( $T_4$ ) concentration (Henneman *et al.* 1955; Annison and Lewis 1959; Sutherland and Irvine 1974). It is not known from these experiments to what extent variations in the composition and amount of feed consumed might have contributed to the  $T_4$  differences observed.

In order to examine whether plasma  $T_4$  concentration changes while feed intake is controlled and whether any changes are related to variations in ambient temperature or to the reported annual cycle of wool growth,  $T_4$  concentrations were measured in plasma samples obtained once weekly throughout a year from 10 non-pregnant sheep maintained on a constant feed intake.

### Materials and Methods

Ten Merino, Border Leicester crossbred castrated male sheep were housed in individual pens in a room well illuminated with daylight from both windows and skylight. No artificial light or temperature control was used. The sheep were shorn once in mid-November. The sheep were fed a daily ration of 800 g lucerne chaff, all of which was consumed. After a settling-in period of 1 month, blood samples were obtained weekly at the same time of day by venipuncture using 200 i.u. heparin per 10 ml blood. Plasma was separated immediately and stored frozen at  $-20^\circ\text{C}$  until assayed.

Daily maximum and minimum inside temperatures were recorded and averaged to give a weekly value.

Plasma  $T_4$  was estimated by a modification of the radioimmunoassay described for triiodothyronine ( $T_3$ ) by Eastman *et al.* (1975). Antiserum against  $T_4$  conjugated to protein was raised in sheep. Unknown sera (10  $\mu\text{l}$ ) were assayed using 26 mg of sodium salicylate per tube to displace  $T_4$  from the thyroxine-binding globulin. No cross-reaction with  $T_3$  was observed over the physiological range. Plasma from thyroidectomized sheep gave zero values when assayed for  $T_4$ . The sensitivity of the assay, defined as the smallest amount of unlabelled  $T_4$  which could be distinguished from no  $T_4$ , was 400 pg/ml. The intra- and interassay coefficients of variation were less than 17%.

All samples from an individual sheep were included in one assay. A standard plasma sample was also included in each assay.  $T_4$  appears to be stable for at least one year in plasma stored frozen at  $-20^\circ\text{C}$  (Wallace, unpublished observations).

## Results

The mean plasma  $T_4$  values and standard errors during a complete year are shown in Fig. 1*a*. Mean weekly maximum and minimum indoor ambient temperatures throughout the same period are shown in Fig. 1*b*. There was a cyclic variation in plasma  $T_4$  concentrations, the highest values occurring in September and the lowest values in April and May. The highest and lowest ambient temperatures occurred in January and June–July respectively.

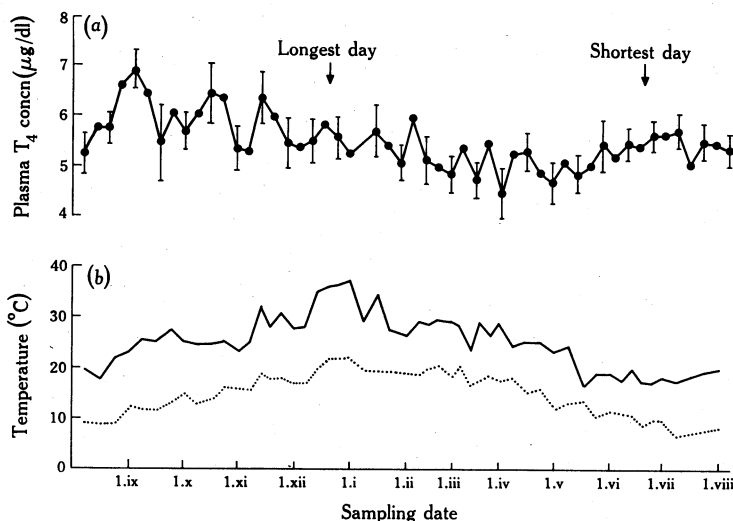


Fig. 1. (a) Total plasma  $T_4$  concentration  $\pm$ s.e. at weekly intervals throughout one year. Each point is the mean value for ten sheep. (b) Maximum (—) and minimum (···) inside ambient temperature. Each point is the mean value of seven readings.

The mean weekly total plasma  $T_4$  concentrations and standard errors for all samples taken from the 10 sheep in the 3-month periods corresponding to the seasons are shown in the following tabulation:

Period	Season	$T_4$ concn ( $\pm$ s.e.) ( $\mu\text{g/dl}$ )
1 (5.ix–27.xi)	Spring	$5.9 \pm 0.1$
2 (4.xii–26.ii)	Summer	$5.3 \pm 0.1$
3 (12.iii–28.v)	Autumn	$5.0 \pm 0.1$
4 (4.vi–28.viii)	Winter	$5.5 \pm 0.1$

The mean for each period was significantly different from the adjacent mean (between periods 1 and 2,  $P < 0.001$ ; 2 and 3,  $0.05 > P > 0.02$ ; 3 and 4,  $P < 0.001$ ; 4 and 1,  $0.01 > P > 0.001$ ). The overall mean was  $5.4 \pm 0.1 \mu\text{g/dl}$ .

## Discussion

In penned sheep maintained indoors under natural light and consuming a constant amount of feed, there were small but significant changes in plasma  $T_4$  concentrations.

The changes were cyclic in pattern, the highest  $T_4$  values occurring in spring and the lowest in autumn. This is in agreement with protein-bound iodine values reported for cattle (Sorensen 1956) where maximum values were found in late spring and minimum values in early autumn. They are also broadly in agreement with the results reported in sheep (Henneman *et al.* 1955; Annison and Lewis 1959; Sutherland and Irvine 1974), although the data in all these papers are few and in the first two only iodine secretion rate and protein-bound iodine respectively were measured. In addition, the measurements were carried out on grazing animals and were confounded by pregnancy and fluctuating feed intake.

Although the animals had been in pens for 1 month before the experimental measurements commenced, it is possible that some of the initial fluctuations in plasma  $T_4$  concentration and the somewhat larger variations seen between animals at the beginning could have been a result of the animals' being confined indoors. It is unlikely that the long term changes were due to this factor.

The energy or protein content of the different batches of lucerne chaff used may have varied, although from past experience such variation is unlikely to have been very large. However, it could have had some effect on plasma  $T_4$  concentrations.

Although the sheep were not pregnant, were on a constant feed intake, and were almost certainly exposed to milder climatic conditions than those in the experiments of Annison and Lewis (1959) and Sutherland and Irvine (1974), the magnitude of the changes in plasma  $T_4$  concentration and in protein-bound iodine were about the same.

The observed changes in plasma  $T_4$  concentration throughout the year did not appear to correlate with reported seasonal changes in wool growth (Hutchinson 1976). Since some seasonal changes in wool growth also occur in thyroidectomized animals maintained with a constant amount of  $T_4$  (Ferguson *et al.* 1965), it is unlikely that  $T_4$  is involved in a causative sense.

In conclusion, the data presented in this paper are the first extensive data for plasma  $T_4$  concentrations in normal, non-pregnant, undisturbed sheep on controlled feed intake. Although it cannot be concluded that changes in climate and day length were major factors in altering plasma  $T_4$  concentrations, nevertheless significant changes throughout the year were observed which were in broad agreement with the changes reported previously in ruminants and which have been attributed to seasonal effects.

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