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Plasma Thyroxine Concentrations in Grazing Sheep in Several Areas of Australia

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Abstract

Plasma thyroxine concentrations were measured by radioimmunoassay in plasma samples from 691 lactating ewes in 26 areas of New South Wales, Queensland, Western Australia and Tasmania. Sheep sampled in New South Wales and Tasmania had significantly lower plasma thyroxine values (4·0 and 4·3 μg/100 ml respectively) than those sampled in Queensland and Western Australia (5·4 and 5·3 μg/100 ml respectively). However, sheep in some districts in southern Queensland also had low plasma thyroxine values.

The areas where sheep had low plasma thyroxine values correlate well with areas where goitre has been previously reported, both in man and in domestic animals. This suggests that measurement of plasma thyroxine is probably a valid empirical method of assessing the relative iodine deficiency of grazing sheep and further that sheep grazing substantial areas of New South Wales, Tasmania and to a lesser extent Queensland may have thyroid dysfunction of varying degrees of severity. These findings could have implications for animal production in these areas.

Introduction

Iodine deficiency can cause goitre, both in grazing animals and in man. The outstanding symptom of its occurrence in grazing animals appears to be reproductive failure (Calderbank 1963). There has been, however, only one reported systematic study in Australia of the thyroid status of grazing animals. In 1932 Dawburn and Farr measured the iodine content of thyroids collected from groups of grazing sheep in several areas of Australia. They concluded that no iodine deficiency existed in Australian pastures. Nevertheless, reports of sporadic outbreaks of goitre in lambs associated with neonatal mortality have appeared from time to time (Walker 1944; Southcott 1945; Clements 1957; Setchell *et al.* 1960; George *et al.* 1966). Also several areas where endemic goitre occurs in humans have been reported (Sutton 1923, 1927, 1933; Clements 1948, 1954; Clements and Wishart 1956).

It would thus be reasonable to conclude, despite the results of Dawburn and Farr (1932), that grazing animals in certain districts of Australia could be deficient in iodine. Identification of such districts requires the measurement of a suitable parameter that can be correlated with iodine deficiency.

There is evidence that plasma thyroxine concentration is related to the level of iodine intake. It has been found by one of us (R.W.M.) that the decrease in plasma thyroxine concentration in ewes fed a low iodine diet during pregnancy could be prevented by the daily administration of 100 μg KI. In addition, ewes fed a wheat

diet low in iodine produced goitrous lambs with significantly lower plasma thyroxine values than lambs born to ewes grazing normal pasture (Hopkins 1972). Also, the injection of iodized oil into humans doubled the plasma thyroxine concentration (Thilly *et al.* 1973).

The present paper describes the results of a survey in which thyroxine concentration was measured in plasma samples obtained from lactating, grazing ewes from a number of properties in various districts of New South Wales, Queensland, Western Australia and Tasmania.

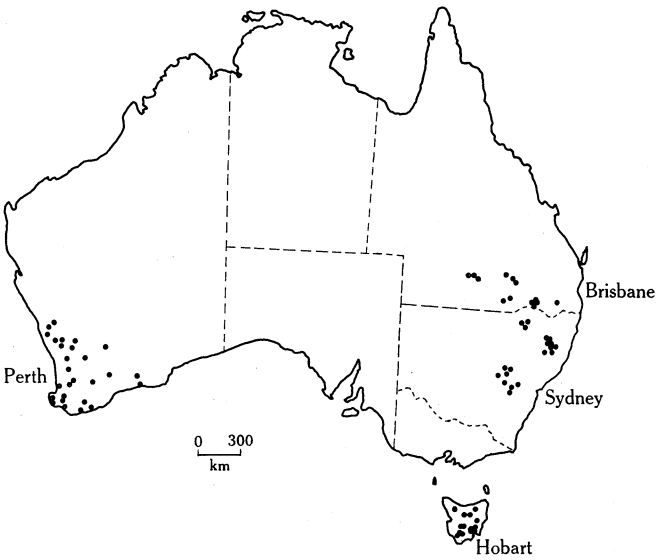

Fig. 1. Areas where sheep were sampled for estimation of plasma thyroxine concentrations.

Materials and Methods

The survey involved 691 lactating ewes on 73 properties from 26 areas of New South Wales, Queensland, Western Australia and Tasmania (Fig. 1). Samples were collected from middle to late lactation and during the period July–November.

Blood (10 ml) was collected into plastic tubes containing 200 units heparin or into unheparinized tubes and the blood allowed to clot. Plasma or serum was separated by centrifugation and transported to the Prospect Laboratory in New South Wales. Neither collection into glass or plastic tubes nor collection as serum or plasma appeared to influence the thyroxine values measured in a particular blood sample.

Thyroxine was measured by the radioimmunoassay method described for triiodothyronine by Eastman *et al.* (1975). Measurements were carried out using 10-μl samples and 25 mg of sodium salicylate was used to displace thyroxine from thyroxine-binding proteins. The antiserum was prepared in sheep by the injection of thyroxine conjugated to protein and emulsified in complete Freund's adjuvant. Triiodothyronine did not interfere with the assay over the range of physiological concentrations.

Samples were randomized between assays and analysed in batches. A standard, control plasma was assayed in quadruplicate in each assay.

An analysis of variance was used to test the significance of the differences in thyroxine values between States, between districts within a State, and between properties within a district.

Results

Four batches of [^{125}I]thyroxine were used during the 4-month period in which the 22 assays were carried out. The use of batches 1 and 2 resulted in higher thyroxine values (4·4 μg/100 ml) for the control standard than did the other two batches (3·3 μg/100 ml). This difference was significant ($P < 0·001$) and the thyroxine values obtained using batches 1 and 2 were corrected using the mean value obtained for the control standard in the other 16 assays. The use of a correction factor made only a small difference to the group means and standard errors since less than 10% of the plasma samples assayed were included in the first six assays.

The mean thyroxine values obtained for the four States are summarized in Table 1. The values for New South Wales and Tasmania were significantly lower ($P < 0·001$) than those for Queensland and Western Australia. Table 2 shows the mean plasma thyroxine values for the groups of sheep sampled on the various properties in the four States. The mean values for the properties are placed in rank order for each district and the statistical differences between districts and between properties within a district are given.

Table 1. Mean plasma thyroxine concentrations in sheep from different States

The comparisons New South Wales *v.* Queensland, New South Wales *v.* Western Australia, Queensland *v.* Tasmania, and Western Australia *v.* Tasmania were all significantly different ($P = 0·01$) using the Duncan multiple range test and ($P < 0·001$) using the least significant difference test. *n*, Number of sheep sampled in each State

State	*n*	Thyroxine concn (μg/100 ml)	s.e.
New South Wales	157	4·0	0·10
Tasmania	140	4·3	0·13
Western Australia	279	5·3	0·09
Queensland	115	5·4	0·21
Totals and means	691	4·8	0·07

Fig. 2 shows the percentage of properties within each State falling within various intervals of thyroxine concentration. Five intervals are shown within the range 2·6–5·0 μg/100 ml with a sixth interval for values greater than 5·0 μg/100 ml.

In New South Wales (Table 2) sheep from the Moree district had significantly higher mean plasma thyroxine concentrations than those from the other three regions ($P < 0·001$). The within-district comparisons for Armidale, Cowra, and Trangie revealed an incidence of about 50% for properties where sheep had plasma thyroxine concentrations of less than 4·0 μg/100 ml. In the Armidale region the lowest values appeared to be associated with light basaltic soils.

Plasma thyroxine values of sheep sampled in Queensland (Table 2) were generally higher than those found in New South Wales. Two properties in Charleville and one property in Roma yielded samples with thyroxine concentrations of less than 4·0 μg/100 ml.

Sheep on half of the 14 properties sampled in Tasmania (Table 2) had plasma thyroxine values below 4·0 μg/100 ml. These properties were fairly well divided among the six districts sampled.

Table 2. Mean (± s.e.) thyroxine (T_4) concentrations in sheep from different properties
n, Number of sheep sampled at each property

District and property	*n*	T_4 (μg/100 ml)	s.e.
New South Wales			
Armidale			
Woolbrook	8	2·8	0·35
Guyra	8	3·3	0·35
Walcha	9	3·4	0·43
Ben Lomond	8	4·1	0·32
Ben Lomond	8	4·2	0·52
Walcha	7	4·2	0·19
Ben Lomond	8	4·5	0·48
Ben Lomond	8	4·7	0·41
Total, means	64	3·9	0·16
Trangie			
Dandaloo	9	2·7	0·27
Trangie	8	3·1	0·40
Trangie	10	4·0	0·53
Trangie	9	4·5	0·34
Total, means	36	4·0	0·10
Cowra			
Cowra	10	3·6	0·25
Eugowra	10	3·8	0·35
Cowra	9	4·4	0·20
Rockley	6	4·8	0·37
Total, means	35	4·1	0·16
Moree			
Wenna	8	4·7	0·30
Wenna	6	4·9	0·46
Moree	8	6·0	0·43
Total, means	22	5·2	0·25
Queensland			
Charleville			
1	10	3·0	0·15
2	5	3·4	0·28
3	13	4·3	0·26
Total, means	28	3·7	0·18
Warwick			
1	13	4·05	0·38
Roma			
1	10	2·9	0·21
2	9	4·5	0·32
3	10	6·7	0·32
Total, means	29	4·7	0·33
Goondiwindi			
1	3	4·7	0·59
2	8	4·9	0·24
3	2	5·3	0·56
4	6	6·8	0·42
Total, means	19	5·5	0·28
St George			
1	15	8·5	0·30
2	11	8·6	0·37
Total, means	26	8·5	0·23
Tasmania			
Derwent Valley			
Ouse	10	2·6	0·21
Hayes	10	3·3	0·28
Plenty	10	3·5	0·27
Ouse	10	4·9	0·66
Total, means	40	3·6	0·23
Huon			
1	10	3·3	0·24
2	10	4·1	0·38
Total, means	20	3·7	0·24
North Midland			
Campbelltown	10	3·6	0·23
Cressy	10	4·3	0·44
Cressy	10	5·2	0·35
Total, means	30	4·3	0·23
North-west coast			
Elliot	10	4·3	0·25
Inner south-east			
Pontville	10	3·8	0·36
Campania	9	3·9	0·33
Richmond	10	7·2	0·36
Total, means	29	5·0	0·36
North-east coast			
Scottsdale	11	5·9	0·41
Western Australia			
Katanning			
Bullock Hills	8	4·1	0·42
Northam			
Beverley	10	4·2	0·22
Minivale	10	4·4	0·25
Grass Valley	10	4·9	0·39
Total, means	30	4·8	0·17
Three Springs			
Three Springs	9	4·1	0·35
Bowgada	9	5·1	0·18
Eneabba	10	5·7	0·52
Total, means	28	5·0	0·25
Albany			
Albany	10	4·9	0·31
Mt Barker	10	5·1	0·44
Denmark	10	5·3	0·51
Total, means	30	5·1	0·23
Moora			
Wannamal	10	4·4	0·23
West Moora	10	5·1	0·43
East Moora	10	5·7	0·54
Total, means	30	5·1	0·25
Manjimup			
Bridgetown	12	5·3	0·51
Manjimup	12	5·9	0·24
Total, means	24	5·6	0·30

Table 2 (*Continued*)

District and property	*n*	T_4 (μg/100 ml)	s.e.	District and property	*n*	T_4 (μg/100 ml)	s.e.
Western Australia				*Western Australia*			
Narrogin				Esperance			
Narrogin	10	3·8	0·23	Salmon Gums	10	4·9	0·32
Williams	10	5·6	0·45	Dalyup North	10	5·6	0·23
Newdegate	10	6·0	0·54	Gibson	9	6·4	0·74
Total, means	30	5·2	0·30	Total, means	29	5·6	0·28
Bunbury				Merredin			
Margaret River	10	4·9	0·27	Moorda	10	4·5	0·23
Cowaramup	10	5·4	0·57	Bruce Rock	10	4·7	0·43
Waterloo	9	5·8	0·71	Southern Creek	11	7·8	0·46
Total, means	29	5·4	0·31	Total, means	31	5·7	0·25

	N.S.W.	Qld	Tas.	W.A.
Differences between districts	$P < 0·001$	$P < 0·001$	$P < 0·001$	$P < 0·001$
Differences between properties within districts	$0·01 < P < 0·05$	Charleville $0·001 < P < 0·01$ Goondiwindi $P = 0·01$ Roma $P < 0·001$	Inner south-east $P < 0·001$ Derwent Valley $0·001 < P < 0·01$ North midland $P = 0·01$	n.s. except: Merredin $P < 0·001$ Narrogin $0·001 < P < 0·01$

In contrast, sheep from only one property (Narrogin) of the 28 sampled in Western Australia had mean plasma thyroxine values of less than 4·0 μg/100 ml. Sheep from 12 properties had plasma thyroxine values of less than 5·0 μg/100 ml and most of these properties were confined to an area east of Perth, bounded by Koorda in the north, Dumbleyang in the south, and Merredin in the east.

Discussion

The measurement of thyroxine in unextracted plasma samples obtained once from grazing ewes during lactation appears to be a useful empirical method for assessing iodine status. Samples were taken during lactation since at this time the ewes were being subjected to a heavy nutritional drain and it was felt that differences in thyroid function would be more easily detected. Although the precise relationship between plasma thyroxine concentration and iodine intake is not known, there is sufficient evidence to show that these are positively correlated. Some of this evidence has been obtained from studies involving the feeding of diets low in iodine to pregnant ewes (R. W. Mason, unpublished data; Hopkins 1972). In addition, Rudert and Oliver (1976) have shown that depression of iodine uptake in pregnant ewes resulted in depressed protein-bound iodine concentrations and goitre in new-born lambs. Iodine added to the diet raised the concentration of protein-bound iodine and prevented goitre. In rats, Greer *et al.* (1975) observed that animals on a low iodine diet showed reduced plasma thyroxine concentrations (<50%) compared to those on an iodine sufficient diet. In humans Thilly *et al.* (1973) showed that plasma thyroxine concentrations rose from a value of 3 μg/100 ml in goitrous subjects to 6 μg/100 ml after the injection of iodized oil to the same subjects.

Although it is not possible at present to define plasma thyroxine concentration limits in sheep on adequate iodine intake, some data are available. Thyroxine concentrations in pregnant ewes have been variously reported to be 5·5 μg/100 ml (Dussault *et al.* 1971), 7·3 μg/100 ml (Thorburn and Hopkins 1973) and 3·8 μg/100 ml (Nathanielsz *et al.* 1973). One of us (A.L.C.W.) has measured thyroxine concentrations in 500 plasma samples taken throughout one year from 10 penned castrated male sheep fed a constant amount of lucerne chaff. Thyroxine values ranged from 4·5 to 6·9 μg/100 ml with a mean of 5·4±0·06 μg/100 ml. In the present survey, many properties yielded values below 4·0 μg/100 ml and it is reasonable to regard these as being possibly iodine deficient.

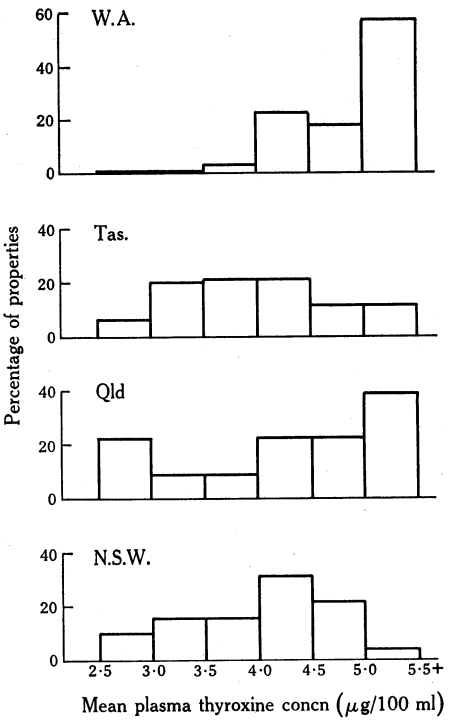

Fig. 2. Percentage of properties in each State falling within various thyroxine concentration intervals.

In New South Wales and Tasmania about 40%, and in Queensland about 30%, of the properties surveyed gave plasma thyroxine values below 4·0 μg/100 ml. In contrast, on only one property in Western Australia was a thyroxine value of less than 4 μg/100 ml obtained and sheep on 16 of the 28 properties had values over 5·0 μg/100 ml.

Western Australia is generally considered to be goitre-free and the above findings support this conclusion. One interesting observation was that 8 of the 12 thyroxine values below 5·0 μg/100 ml came from sheep in a fairly well-defined area east of Perth.

In New South Wales goitre has been reported in lambs from the districts of Crookwell (Setchell *et al.* 1960), Armidale and Glen Innes (George *et al.* 1966) and Monaro (Anon. 1970). We found low levels of thyroxine in sheep from the Armidale district and in areas around Trangie, Parkes and Cowra. Goitre has also been observed in humans in New South Wales. Sutton (1933) reported a high incidence of goitre

along the edges of the New England tableland and, in particular, to the west of Armidale. More recently, a study of the incidence of goitre in school children by Hales *et al.* (1967) suggests that there is still a high incidence of goitre in many areas of New South Wales. The only relationship between soil type and thyroxine concentration was that ewes grazing pasture on the light basaltic soils around Walcha and Woolbrook had lower thyroxine levels than those on the heavier basalt soil around Ben Lomond.

Clements (1957) reported goitre in school children around Warwick in southern Queensland and noted that the incidence varied from district to district. Our observations of low thyroxine values in sheep from several districts of southern Queensland suggest that endemic goitre may be more widespread than has been suspected.

Practically all of Tasmania is considered an endemic goitre area for humans, with the highest incidence in the southern part of the island (Sutton 1933; Clements 1954; Clements and Wishart 1956; Clements 1957).

There have been several reports of outbreaks of goitre in domestic animals in Tasmania (Southcott 1945; Green 1956; Clements 1957). More recently Statham and Bray (1975) and one of us (R.W.M.) have found that ovine congenital goitre is regionally distributed. This pattern of distribution is allied to the three main river systems—the Huon, the Derwent, and the confluence of the South Esk, Lake and Macquarie Rivers. In addition, Statham and Bray (1975) have shown that pasture grown in sandy soils contains less iodine than pasture grown in clay soils. This relationship between soil type and plant iodine is supported by the demonstration that milk iodine levels are lower in ewes grazing pastures on sandy soils than in those grazing pastures on clay soils (R. W. Mason, unpublished data). Also, the development of ovine goitre occurs mainly in sheep grazing sandy soils in these areas.

If the thyroxine levels are statistically analysed in accordance with the regional distribution of goitre, then sheep sampled from the three goitre regions described—Huon, Derwent Valley and Cressy–Longford—all had significantly lower thyroxine levels when tested by the method of least significant differences ($P < 0{\cdot}001$, $P < 0{\cdot}001$ and $0{\cdot}01 < P < 0{\cdot}05$ respectively) than the combined thyroxine levels from the remainder of the State. Most of the sheep sampled in the southern half of the State also had low thyroxine values.

Apart from low iodine intake due to low iodine content of pastures, low plasma thyroxine values could also arise from the presence of goitrogens in particular pasture species, e.g. white clover (George *et al.* 1966). However, to date there is no unequivocal evidence that pasture goitrogens are a primary cause of endemic goitre (Trikojus 1974).

It can be concluded that in all States assessment of the distribution of iodine-deficient areas on the basis of plasma thyroxine concentrations agrees in general with previous assessments based on the incidence of goitre. In Queensland, at least, the findings suggest that iodine deficiency may be more widespread than hitherto suspected.

Some of the variation between States could be due to different susceptibilities of different breeds to goitre. No information is available on this point apart from the report of George *et al.* (1966) who showed different incidences of goitre in Dorset Horn and Merino sheep.

More measurements are necessary to define more precisely the incidence and distribution of iodine-deficient areas in districts where low thyroxine values have been found. In particular, changes from year to year in these areas need to be assessed.

Also the significance of these findings in relation to losses in animal production remains to be investigated.

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