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S T U D I E S

O N   T H E   T H Y R O I D   G L A N D

O F   C A T T L E

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INTRODUCTION

The health and production of the dairy cow is dependent upon the efficient functioning of many interrelated physiological processes. For many years considerable attention has been paid to digestion, absorption of nutrients, circulation of the blood, utilisation of metabolic products, and excretion. It is only in recent years, however, that the importance of the endocrine system has been appreciated, and there still remains a large amount of fundamental research to be carried out before a complete understanding of the role of hormones will be obtained.

In the endocrine system, the thyroid gland is a most important unit in that its efficient functioning is essential to normal growth, metabolism and lactation. Thyroidectomy and hypothyroidism have been shown to depress body growth and to lower basal metabolic rate. Hyperthyroidism on the other hand, causes an increase in heart rate and oxygen uptake, a loss of nitrogen, emaciation and hyperirritability. That the thyroid plays an important role in the lactating dairy cow has been demonstrated by experimental work - hypothyroidism has been shown by Graham (1934) and Spielman, Petersen and Fitch (1943) to cause a decrease in milk and fat production, while Graham (1934) and many later workers have shown that mild hyperthyroidism may cause an increase in both the milk yield and the butterfat content of the milk.

In the past seven years, there have been two developments which are facilitating studies of thyroid function. Firstly, Ludwig and von Mutzenbecher (1939) have provided a method for the preparation of cheap, thyroid-active iodinated proteins. With such materials it is possible to supplement the hormone output of the thyroid gland of the animal at will. Secondly, a means of producing hypothyroidism without thyroidectomy has been developed from the work of Mackenzie, Mackenzie and McCollum (1941) and Kennedy (1942) who discovered the goitrogenic nature of sulphonamides and thioureas.

These two experimental aids were used in the projects reported in this thesis, where the following problems were studied.

As iodinated proteins may soon be used in farming practice to stimulate milk and butterfat production, it is important to know the effects of such materials on all the organs of the dairy cow. In this thesis are recorded preliminary experiments which were undertaken to study the effect of thyroid-active iodinated casein (thyroprotein) on the thyroid gland.

Goitrogenic substances, such as thiourea and thiouracil, are useful in studying the part played by the thyroid gland in the lactation process in that they provide a means of observing, without surgical intervention, the effect of thyroid deficiency on lactation. One of the first problems in applying this technique to dairy cattle is the determination of a dosage of a goitrogen which will definitely affect thyroid activity in these animals without being too toxic for practical use. To determine such an effective dose rate, a study has been made of the effect of various dosages of thiourea upon the thyroid gland of dairy cows, and the results of the project are recorded in this manuscript.

Goitrogens have the effect of inhibiting the formation of thyroxine by the thyroid gland. This results in a compensatory hypertrophy and hyperplasia of the thyroid due to an increased output of the thyrotrophic hormone by the anterior pituitary gland in response to the induced thyroid deficiency. By the administration of thyroxine, dessicated thyroid gland, or artificial thyroprotein, the pituitary hormone can be held in check and the effect on the thyroid of goitrogen-treated animals reduced in proportion to the dosage of thyroidal substance. The normal thyroid hormone output can be estimated by determinigg the amount of thyroxine required to return the thyroid glands of goitrogen-treated animals to normal. The work reported in this thesis is fundamental in the application of this technique to dairy cattle where it possibly may be used to determine whether the thyroid activity of high producing cows differs markedly from that of low producers.

Because very little data are available at the present time concerning the size of the thyroid gland in New Zealand cattle, the weights of thyroid glands obtained from cattle slaughtered at freezing works and abattoirs in the Manawatu are presented. It is realised that these data are inadequate to give a comprehensive picture of the thyroid weights of New Zealdnd cattle; the data shown must be regarded as those of a preliminary study only.

THE ANATOMY OF THE THYROID GLAND

REVIEW OF LITERATURE

General Anatomy of the Thyroid Gland

The thyroid gland of most animals is composed of two lateral lobes connected by an isthmus and is located in close association with the anterior and lateral parts of the trachea, at the base of the larynx. It is a very vascular ductless gland, firm in texture and pale red in colour. The two lateral lobes are irregular in outline, and extend along either side of the larynx as conical or pyramidal projections. Accessory thyroid tissue is reported as occurring frequently (Wohl, 1917; Crotti, 1938) making total thyroidectomy difficult to perform.

The gland is enclosed by a thin connective tissue capsule which is connected with the surrounding cervical fascia, and which also projects inwards to divide the gland into lobules.

The tissue of the gland is compact, and consists of lobules which are embedded in a stroma of fibrous tissue. The stroma contains numerous blood and lymph vessels. The lobules consist of non-communicating alveoli or follicles of varying size and form. The follicle are lined by cuboidal or columnar epithelium, and the lumina usually contain a characteristic viscid material, called colloid. Some follicles may be without lumen, and consequently do not contain colloid.

The amount of colloid found in a gland is by no means a true index of the functional activity of the thyroid; the amount present depends upon the two processes of secretion into the lumina and absorption from the lumina into the bloodstream, and the influence of the thyroid on metabolism depends largely upon the equilibrium between the processes. For example, if there is a rapid absorption of thyroid products from an extremely active gland, little or no colloid will be found. On the other hand, the quantity of colloid may be found to be materially increased in cases in which secretion and absorption are diminished. In these two instances false conclusions as to the functional activity of the gland would be drawn if one were to assume that activity is directly proportional to the amount of colloid present.

The follicle epithelium of the gland shows great variations in size and arrangement of cells, influencing factors being age, sex, season of the year, and diet. In general, it is believed that the epithelium becomes very low when the secretory activity of the gland is depressed, and very tall and folded when the gland is hyperactive.

The anatomy of the thyroid gland in cattle agrees well with the outline of general anatomy of the thyroid given above.

The Weight of the Thyroid Gland in Cattle

The weight, size and activity of the thyroid gland has been shown to be affected by many factors. Keating and others (1935) have shown that thyroid weight is increased by both colloid accumulation and increases in epithelial constituents. Thyroid weight as an index of thyroid activity is further complicated by the fact that variation in the weight of the gland may be due to vascularity and stroma apart from changes in the amount of colloid and number of acinar cells. A marked increase in the weight of the gland, however, may be taken as indicative of thyroid dysfunction.

Krizenecky (1932) has tabulated data from the work of Trautman (Drusen mit innerer Secketion-Joasts Spegille patholog-ische anatome der Haustier (1917). These data present a comparison of thyroid gland measurements between dairy cattle and other domestic animals, but no account was taken of differences due to breed.

TABLE I

Comparative Measurements of Thyroid Glands of Domestic Animals (Trautman, 1917)

	Thyroid length (cm.)	Thyroid width (cm.)	Thyroid thickness (cm.)	Thyroid weight (gm.)
Dairy cattle	6.5-7.5	4.00-5.0	0.85-1.50	23-41
Beef cattle	6.0-7.0	4.00-5.0	0.75-1.50	21-36
Horses	3.5-4.0	2.3	1.50	20-35
Swine	4.0-4.5	2.00-2.5	0.50-0.75	12-30
Sheep	3.0-4.0	1.25-1.5	0.50-0.75	4-7
Goats	2.5-5.0	1.00-1.5	0.50-0.80	8-11

Krupski (1921) reported on the size of the thyroid in dairy and beef cattle of various ages and conditions from which Krizenecky (1932) tabulated the data shown in Table II. In this work, too, no data was presented to show differences between breeds.

Swett, Matthews, Millar and Graves (1937) have presented data showing a difference in weight of thyroid between breeds of dairy cattle. A summary of these data is given in Table III.

TABLE II

Thyroid Weights in Cattle (Krupski, 1921)

Type of Animal	Number of animals	Age (years)	Absolute weight (gm.)
Beef cattle	40	1-5	21.49
Beef cows (pregnant)	21	1-4	21.76
Steers	67	1-5	42.08
Milk cows	171	2-13	32.59
Milk cows (pregnant)	69	3-11	31.80

TABLE III

Average Thyroid Weights of Dairy Cows Compared with Body Weights (Swett, Matthews, Miller and Graves, 1937)

	Number of animals	Live weight (lb.)	Empty body weight (lb.)	Thyroid weight (gm.)
Ayrshires	44	1083	931.98	33.4
Guernseys	62	991	859.62	31.6
Holsteins	198	1265	1089.59	38.1
Jerseys	218	910	788.56	27.9

EXPERIMENTAL

Methods

In order to collect data concerning the size of the thyroid gland in cattle in New Zealand, periodic visits were made to the freezing works at Longburn and Feilding, and to the Palmerston North Municipal Abattoirs. As much information as possible about the previous history of the animals was obtained from the butchers concerned.

It was planned to study possible differences in thyroid weight due to age, weight, sex, and breed of the animal, function, i.e. beef and dairy breeds, locality and season of the year. The number of animals of dairy breeds from which glands could be obtained was comparatively small, and so it is not possible at this intermediate stage of the survey to analyse the effects of factors such as age, sex, locality and season in the case of dairy cattle. These factors, however, have been analysed in the case of beef cattle, and with the continuation of the project it is hoped to accumulate sufficient data for dairy cattle. Carcass weights were largely unobtainable in the survey on account of the organisation of freezing works. Those dairy cows from which thyroid glands were obtained were in all cases either dry animals or strippers.

A major difficulty was to obtain the glands intact, for unfortunately it is a common practice of the butchers to sever the gland when the head of the animal is being removed. A certain percentage of the glands were not severed however, and by obtaining co-operation on the part of some of the butchers, the difficulty was overcome to some extent. Only intact glands are included in the data presented.

The glands were dissected from the tracheas of the animals immediately after the heads had been removed from the carcasses, and the thyroids were then trimmed of all adhering fat and connective tissue and immediately weighed.

Results

A summary of the mean thyroid weights obtained to date for the various classes of cattle is given in Table IV. Full information on the weights of the individual thyroids is given in Appendices I to V.

TABLE IV

Mean Thyroid Weights

Type of animal	No. of animals	Mean thyroid weights (gm.)	Notes
Dairy calves (Jersey) Males	33	7.4±2.39 *	Collected from "Bobby" calves at Longburn works
Dairy calves (Jersey) Females	42	7.6±2.45	
Dairy Calves (Jersey) Males	7	9.7±1.19	Collected from calves killed at birth at Massey College
Dairy cows Jersey	81	29.5±8.64	
Dairy cows Ayrshire	20	34.2±5.69	
Dairy cows Friesian	8	42.0±12.60	
Dairy bulls Jersey	2	34.9±13.7	
Dairy bulls Friesian	2	45.3±3.1	
Vealers (Aberdeen Angus) Castrated males	17	16.4±3.20	
Vealers (Aberdeen Angus) Females	7	17.6±5.32	
Beef heifers (Aberdeen Angus)	36	27.7±7.56	
Beef cows (AberdeenAngus)	15	31.8±4.51	
Steers (Aberdeen Angus and A.A. x Hereford)	87	29.1±7.29	

(* The standard deviation is shown after the mean thyroid weight)

In order to test the significance of the differences in the thyroid weights of the three breeds of dairy cattle shown above an analysis of variance (Snedecor, 1940, p.179) was applied to the data. The results are shown in Tables V and VI.

TABLE V

Analysis of Variance of Thyroid Weights Between Dairy Breeds

Source	d.f.	Sum of squares	Mean square
Total	108	9604.87	
Variance between breeds	2	1341.70	670.86
Variance within breeds	106	8263.17	77.95

F = 8.60 highly significant

The analysis shows a highly significant variation in thyroid weight between breeds of dairy cows. To test the significance of the difference in mean thyroid weight between breeds, "t" tests were applied (Snedecor, 1940, p.57) and the results shown in Table VI.

TABLE VI

Tests of Significance of Difference in Mean Thyroid Weight Between Dairy Breeds

Groups compared	"t"	Significance
Jerseys and Friesians	3.83	Highly significant
Jerseys and Ayrshires	2.14	Significant
Friesians and Ayrshires	2.11	Significant

The analysis shows that the difference in mean thyroid weight between the Jersey, Ayrshire and Friesian cows was statistically significant. This probably was a reflection of different body weights. These results are in agreement with the findings of Swett et al. (1937) in dairy cattle in the United States.

The effect of sex on the thyroid weight was examined between the means of various groups of animals, as shown in Table VII, using the "t" test of significance.

TABLE VII

The Effect of Sex on the Weight of the Thyroid Gland

Groups compared	"t"	Significance
Male and female Jersey calves	0.112	Non-significant
Castrated male and intact female Aberdeen Angus vealers	0.750	Non-significant
Aberdeen Angus cows and A.A. steers	0.440	Non-significant

These results support the conclusion that sex has no effect on the weight of the thyroid gland of male and female Jersey calves. No significant difference was found between the thyroid weights of castrated male and intact female Aberdeen Angus vealers, and cows and steers of the Aberdeen Angus breed.

The number of thyroid glands collected from dairy bulls was too small to permit an analysis of difference in thyroid weight due to sex in mature dairy cattle.

To examine the effect of age on the thyroid weight of Jersey cattle, the data was divided into age groups which included glands from bobby calves, and from cows of 3, 4, 5 and 6 or more years of age. The mean of each group was plotted on a graph as shown in Figure I. The difference between the mean of the group

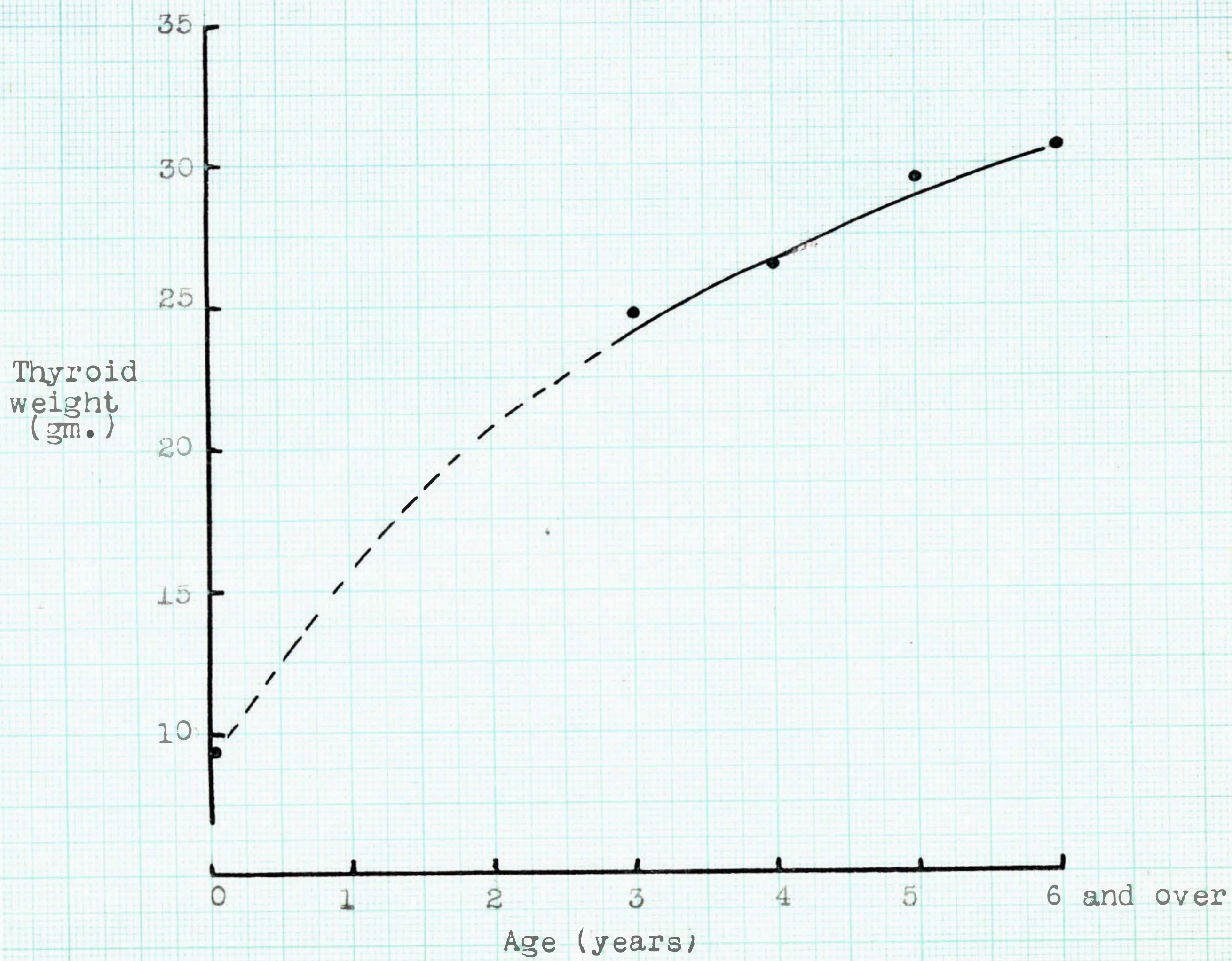


FIGURE I

The Relation Between Thyroid Weight and Age of the Animal in Jersey Dairy Cows consisting of 3 year old animals and that of the group of animals of 6 or more years of age was found to be statistically non-significant (t=1.768), and so further analysis of the data in this respect was not pursued in the meantime.

The difference between the means of the thyroid weights of beef heifers and beef cows was significant (t=2.32), and an increase in thyroid weight with age was thus shown.

To investigate a possible difference in thyroid weight due to function of the animal, i.e. whether used for beef or for dairy production, a "t" test was applied to the difference between the mean thyroid weight of Aberdeen Angus cows on the one hand, and that of Jersey, Ayrshire and Friesian thyroid weights respectively, on the other. The results of these analyses are shown in Table VIII.

TABLE VIII

Effect of Function of the Animal on the Weight of the Thyroid Gland

Groups compared	"t"	Significance
Aberdeen Angus and Jersey cows	0.317	Non-significant
Aberdeen Angus and Friesian cows	2.821	Significant
Aberdeen Angus and Ayrshire cows	1.347	Non-significant

The average body weight of the beef cows from which the thyroids were obtained was about 1300 lb., and the average body weight of the Friesian cows was about 1250 lb., the Jersey cows, 950 lb., and the Ayrshires, 1100 lb.. In view of the results shown in Table VIII, therefore, the Aberdeen Angus cows had a lower thyroid weight per unit of body weight than did animals of the dairy breeds.

Analysis of the data in respect to locality was limited by the fact that the majority of the animals came from the Kairanga district. In two cases, however, a comparison between animals from different districts could be made. Forty-one of the steers came from Hunterville, and when the mean of the thyroid weights of these animals was compared with that of steers from the Kairanga, the difference between the means was found to be statistically non-significant (t=0.270). The calves that were killed at birth at Massey College had thyroids with a mean weight 2.3 grams heavier than those of male calf thyroids from animals off the Kairanga plains. This difference between the means was found to be statistically non-significant (t=0.250).

In the course of a few years it is hoped to collect thyroid glands from each group of cattle at each of the four seasons of the year, and so to determine the effect of season on the thyroid weight. However, because the number of animals slaughtered, apart from "bobby" calves, in the spring months is small, some time will necessarily elapse before such an analysis can be made.

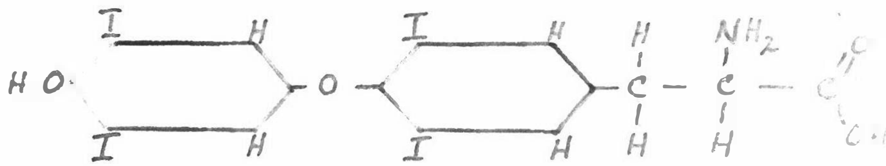
THE FUNCTIONING OF THE THYROID GLAND

REVIEW OF LITERATURE

Chemistry of the Thyroid Hormone

The thyroid provides, through its specific hormone, a means for varying the rate of metabolism to meet changing physiological needs.

A voluminous literature on the chemistry of the thyroid secretion has appeared within the last 45 years. Kendall, in 1914, isolated a crystalline substance with physiological activity from thyroid material but it was left for Harington (1926) and Harington and Barger (1927) to determine the correct chemical formula for the active principle thyroxine, as shown below:-



Thyroxine

Kendall (1929) showed that upon alkaline hydrolysis thyroid tissue is divided into an acid-soluble and acid-insoluble fraction. The acid-insoluble fraction contains thyroxine and possibly other iodine-containing compounds. Harington and Randall (1929) isolated di-iodotyrosine from the thyroid, and Harington (1944) produces impressive evidence, both chemical and biochemical, for the assumption that thyroxine is in fact formed in the body from tyrosine and iodide through the intermediate stage of di-iodotyrosine. Schachner, Franklin and Chaikoff (1943) showed that the biological process is in part controlled by an oxidising enzyme system. The relative amounts of thyroxine and di-iodotyrosine in the thyroid varies quite markedly with the season of the year (Harington, 1933).

The Relation of Iodine to the Thyroid Gland

It is established that iodine is a normal constituent of the thyroid gland and must be supplied to an animal for normal activity of the gland. The thyroid has a marked affinity for iodine as shown by the high proportion of dietary iodine found in the thyroid glands of animals on diets which contain very small traces of iodine. Burk and Grummitt (1945) review experiments in which the passage of radio-active iodine was traced from the diet to the thyroid gland where the iodine was quickly concentrated in large quantities relative to the amount fed. Marine and Rogoff (1916) reported that the iodine content of the thyroid of the dog may be increased several hundred per cent within five minutes following an injection of 50 mg. of potassium iodide intravenously. Eighteen per cent of the iodine contained in a dose of 38 mg. of potassium iodide was recovered by the thyroid. Van Dyke (1925) showed that free iodine and sodium iodide is much more readily taken up, Thyroxine has but little effect on the iodine content of the gland (Ralston et al., 1940).

Koch (1913) first reported a seasonal variation in the iodine content of the thyroid, and Seidell and Fenger (1913), Fenger (1918), and Kendall and Simonsen (1928) confirmed and extended this work. The glands contained less iodine in the winter months and more in the summer months. Salter (1940) presents evidence for a greater secretion of thyroid hormone into the blood stream in cold weather in that there is a reduced storage of colloid by the gland, and a decreased iodine content, in winter.

Regulation of Thyroid Activity

It has long been suspected that a very efficient mechanism exists for regulating thyroid activity by way of the bloodstream. First proof of a pituitary hormone being the regulating factor, however, followed the demonstration by Smith (1917) and by Allen (1919) that hypophysectomy caused atrophy of the thyroid, and Smith's later demonstration (1926, 1930) that implants of fresh pituitary restored such atrophic thyroids to normal and even increased activity. These results have since been confirmed by many workers. The pituitary factor concerned has been called the thyrotrophic hormone, or thyrotrophin.

While the thyrotrophic hormone of the anterior pituitary appears to be the means of stimulating the thyroid, however, there is ample evidence also that the thyroid secretion exercises an influence on the anterior pituitary, particularly in respect to control of the production and release of the thyrotrophic hormone.

The activity of the thyroid and that of the pituitary gland are interdependent, the one regulating the other. Any deficiency in the thyroid secretion quickly stimulates the pituitary, either directly or indirectly, to produce more thyrotrophic hormone; and conversely, an excess of thyroid secretion tends to reduce the secretion of thyrotrophin by the anterior pituitary gland.

Physiologically Active Iodinated Proteins

(a) Discovery. Oswald (1910) found that iodination of albumin, gliadin and casein in alkaline solution gave products from which he was able to isolate 3:5 di-iodotyrosine; this drew attention to the importance of tyrosine as an iodine-containing constituent of the iodinated protein molecule. Abelin (1933, 1934, 1936 and 1938) claimed to have obtained physiologically active iodinated proteins from which, by alkaline hydrolysis, he could separate fractions having the general physical and chemical properties of thyroxine; in only one instance, however, was a crystalline substance having physiological activity isolated and this was not satisfactorily characterised.

Ludwig and von Mutzenbecher (1939) published a detailed description of a method for the iodination of casein and other proteins to give products which were not only physiologically active themselves but from which it was possible to isolate thyroxine. This work was confirmed by Harington and Pitt-Rivers (1939).

The above work was important in that it offered the possibility of cheap and easy preparation in large quantities of products having the specific physiological products of the thyroid gland. This possibility was realised by Reineke and Turner (1942a) who confirmed the original observation, extended the work to other proteins, and defined more sharply the conditions to be observed if optimal yields of active material are to be obtained.

The thyroxine formed from di-iodotyrosine (itself prepared from natural 1-thyroxine) is laevo-rotatory, and therefore identical with natural thyroxine isolated from the thyroid by enzymic hydrolysis.

(b) Physiological Effects of Artificial Thyroproteins.
The effects of hypothyroidism are analogous in all mammals and in

general are more severe the earlier in life the thyroid function is impaired. The effects include depressed body growth; lowered basal metabolic rate; decreased nitrogen retention; carbohydrate absorption from the intestine is lowered; and fat tends to be stored. Good reviews are given by Kojimi (1917), Hoskins (1927) and Salmon (1938).

It is widely recognised that the symptoms of hypothyroidism can be alleviated by the administration of dessicated thyroid gland, thyroxine, or various thyroid-active preparations. This has been shown by Pick and Pineles (1906), Basinger (1916), Smith et al. (1927), Salmon (1938), Evans et al. (1939), and Lerman and Salter (1939). It has been demonstrated by Reineke and Turner (1942a) that artificial thyroproteins may also be used successfully to alleviate the effects of hypothyroidism.

It was first shown by Reineke and Turner (1942a, 1942b) that artificial thyroprotein, when fed to cows and goats in declining lactation, has an effect similar to that of thyroid material or thyroxine, in stimulating increased yields of milk and butterfat. This work has been confirmed and extended with cows lactating normally by Reineke (1943), Van Landringham et al. (1944), Reece (1944), Seath et al. (1944), and Blaxter (1943, 1945a, 1945b, 1946), and with cows brought into lactation with diethylstilbestrol by Hollard and Campbell (1946).

It has been shown by several of the above workers that the increases in milk yield and butterfat obtained by the administration of iodinated proteins to dairy cows is accompanied by an increase in heart rate and a corresponding decrease in body weight.

More information is required, however, on further physiological effects of feeding iodinated proteins. For example, what is the effect on the cow of feeding thyroprotein over a period of several months? What is the effect on longevity of the animal if the feeding is continued over several lactations? What is the effect of the thyroprotein on the organs concerned with the secretion of milk? What is the effect of feeding thyroprotein on the thyroid gland of the animal?

Very little investigation has apparently been carried out on the effect of thyroid material or thyroxine on the thyroid gland. However, the fact that rats fed dessicated thyroid gland show signs of marked functional depression of their own thyroid glands has been demonstrated by Abel et al. (1925), Cordonnier (1929), Reforzo-Membrives (1938), Uotila (1940), Belasco and Murlin (1941), Galli-Mainini (1941), and Koger and Turner (1943). Reforzo=Membrives (1938) reported that the pituitary glands of such rats have a thyroid-inhibiting action when extracts are injected into guinea pigs. An excess of thyroid hormone in the circulation as a result of the administration of dried thyroid may act upon the mechanism in three ways - (1) directly causing an inhibition of the thyroid (Uotila (1940); Galli-Mainini (1941)); (2) inhibiting the thyroid-stimulating action of the anterior pituitary gland (Reforzo-Membrives (1938)); or (3) originating or developing an active thyroid-inhibiting action in the anterior pituitary gland (Reforzo-Membrives (1943)

EXPERIMENTAL

The experiments recorded in this manuscript were undertaken in order to study the effect of thyroprotein on the thyroid gland of the dairy cow.

The thyroprotein used was prepared, using dairy factory equipment, by Mr. I. D. Morton and Dr. I. L. Campbell of the Dairy Research Institute (N.Z.), according to the method of Reineke and Turner (1942), with minor modifications.

Potency of Thyroprotein

To obtain some measure of the potency of the thyroprotein, the material was fed to two dairy cows in declining lactation, and the effects noted on the yield and composition of the milk, and the body weights and pulse rates of the animals recorded. Details of the cows used are shown in Table IX.

The thyroprotein was fed to the Friesian cow, Robbie, at dosages varying from 20 to 45 grams daily over periods ranging from 8 to 56 days. The Ayrshire cow, Sulu, received thyroprotein over only one experimental period, when she received 30 grams daily over 14 days. The method of administering the material was to suspend the thyroprotein in a pint of warm water with approximately one gram sodium bicarbonate, and the suspension was given to the animals as a drench. A definite increase in milk yield was observed about three days after the beginning of the feeding period, and an increase in the butterfat test from about the seventh day. These effects on milk yield and composition were accompanied by a marked rise in the heart rates and a rapid decrease in the body weight of the animals. The results of these trials are shown graphically in Figs. II and III.

The results indicate that the thyroprotein used was slighly less potent than the synthetic thyroprotein "Protamone",

TABLE IX

Details of Cows Used to Estimate the Potency of the Thyroprotein

Name	Breed	Age at beginning of experimental period (years)	Date dried off from last lactation	Date commencing present lactation	Days in milk at beginning of experimental period	Pregnancy status	Notes
Robbie	Purebred Friesian	6	27-7-42	28-12-43	60	Empty	Brought into lactation with diethylstilbestrol
Sulu	Purebred Ayrshire	10	26-5-45	14-9-45	244	Empty	-

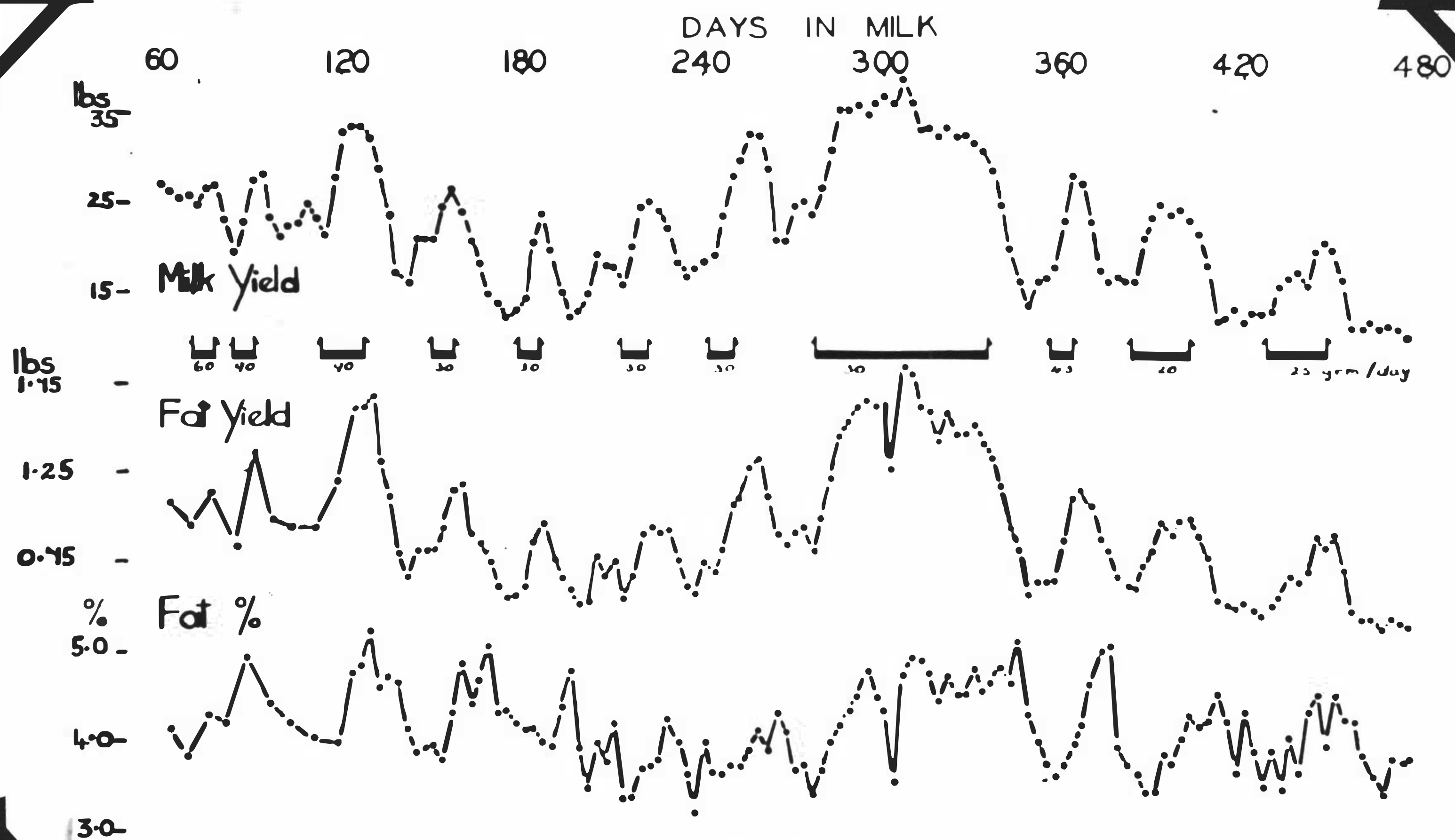
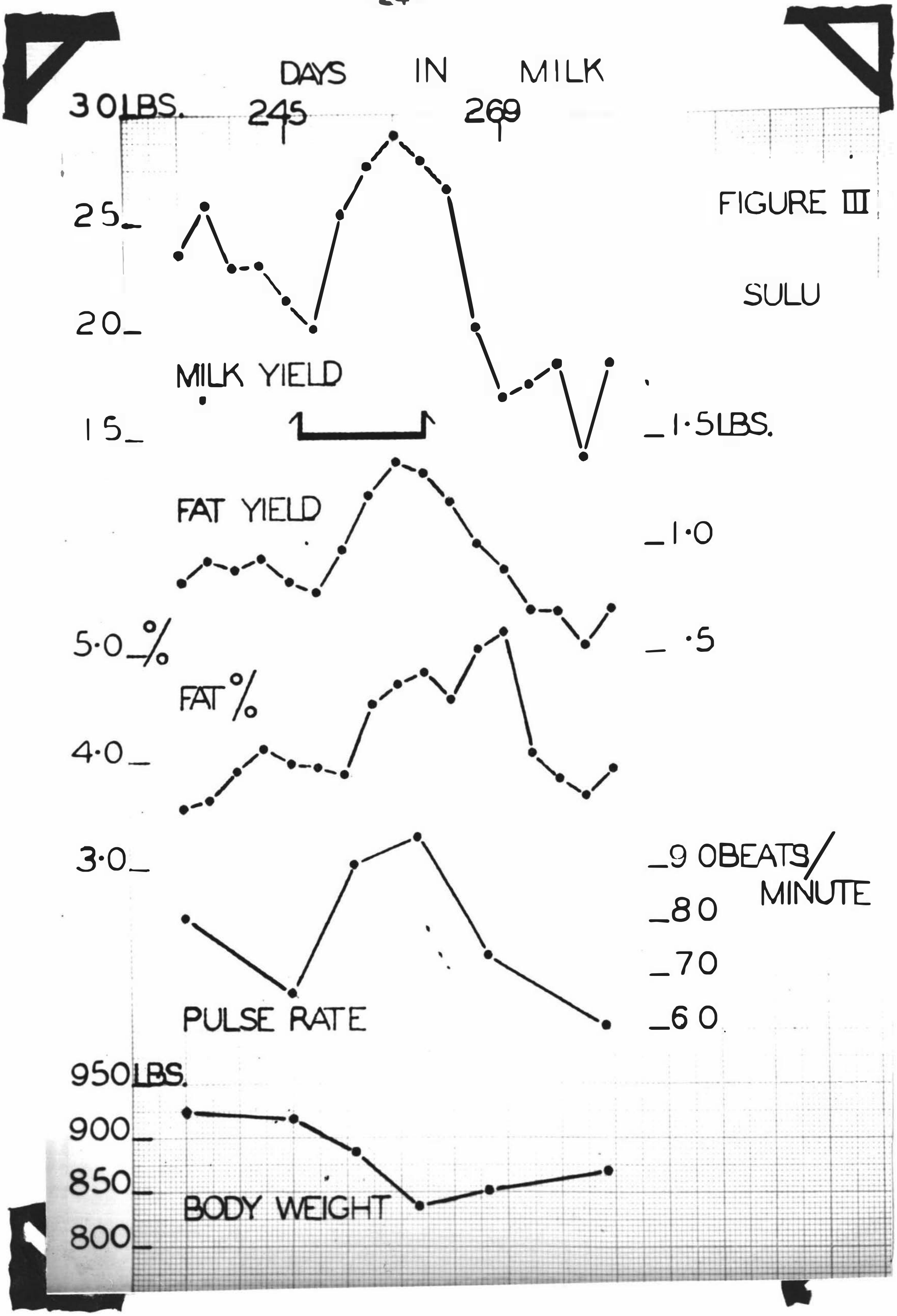


FIGURE 11.

Effect of Thyroprotein on Yield and Composition of Milk

Lactation curves of the Friesian cow "Robbie", which was fed tyyro-
protein at intervals throughout lactation.



Effect of Thyroprotein on Yield and Fat Content of Milk, Pulse Rate and Body Weight.

Results from Ayrshire cow "SULU", fed 30gm. thyroprotein daily over 14 days.

made by Cerophyl Laboratories, Kansas City, Missouri, and used by Reece (1944), Seath et al. (1944), and Van Landringham et al. (1944), and that it was very similar in potency to the mixture of a large number of preparations of iodinated casein used by Blaxter (1946).

In order to study the effect of thyroprotein on the thyroid gland of dairy cows, the material was fed in the above manner at varying dosages to dry dairy cows. Details of the animals and dose rates used are shown in Table X. In order to provide a comparison with the thyroids of animals treated with thyroprotein, thyroids of eleven normal cows were examined histologically. Details of these animals are shown in Table X.

General and Histological Technique

During the experimental period the animals were weighed and their pulse rates determined on three consecutive days weekly. Heart rates were recorded by palpating the saphenous artery on the medio-lateral surface of the hind leg. The animals were allowed to stand quietly for five minutes prior to the observation, and then recordings were made until a constant figure was obtained. Heart rates were not recorded in all cases because the refractory nature of some of the animals used made accurate determinations impossible.

On the day after the conclusion of the feeding period the animals were slaughtered. The thyroids were dissected out immediately after slaughter, trimmed of fat and connective tissue, and were weighed. A sample of tissue was taken from each gland for histological examination by snipping out a small portion of

TABLE X

Details of Cows Used

Name	Breed	Age at killing (years)	Last drying off date	Date of killing	Pregnancy status when killed	Reason for culling	Notes	Treatment in present experiment
A. Normal Cows								
Tramp	Grade Jersey	5	27-5-43	1-3-45	Empty	Empty. Mastitis infection	-	Normal control cow
Tiki	Purebred Jersey	5	12-5-45	22-5-45	Empty	Low producer. Poor type	-	Normal control cow
Phlox	Purebred Jersey	8	29-4-45	31-5-45	Empty	Empty. Unsound in udder	-	Normal control cow
Squire	Grade Jersey	5	27-5-43	21-8-44	Empty	Sterile	-	Normal control cow
Larkspur	Purebred Jersey	11	12-5-44	9-6-44	Empty	Old age	-	Normal control cow
Quartz	Jersey-Ayrshire X	7	13-5-44	9-6-44	Empty	Empty Mastitis infection	-	Normal control cow
No. 16	Ayrshire	5	24-4-44	20-10-44	Empty)	Animals culled from Ruakura herd and sent to Dairy Research Institute where they were used as normal control cows		Normal control cow
No. 19	Ayrshire	5	24-4-44	20-10-44	Empty)			Normal control cow
No. 51	Ayrshire-Shorthorn X	4	24-4-44	20-10-44	Empty)			Normal control cow
No. 67	Shorthorn Jersey X	9	7-3-44	20-10-44	Empty)			Normal control cow
No. 84	Ayrshire Shorthorn X	6	23-4-44	20-10-44	Empty)			Normal control cow

TABLE X (Cont'd.)

Name	Breed	Age at killing (years)	Last drying off date	Date of killing	Pregnancy status when killed	Reason for culling	Notes	Treatment in present experiment
B. Cows Fed Thyroprotein								
Mona	Grade Jersey	3	-	3-7-46	Empty	Sterile	-	10 gm. thyroprotein daily over 14 days
Nell	Grade Jersey	3	-	29-6-46	Empty	Sterile	-	10 gm. thyroprotein daily over 14 days
Mandolin	Purebred Friesian	8½	7-1-45	22-2-45	Empty	Falling production. Mastitis infection	-	20 gm. thyroprotein daily over 14 days
Quiver	Jersey & Jersy-Ayrshire	7½	1-1-45	22-2-45	Empty	Mastitis infection	-	20 gm. thyroprotein daily over 14 days
Lilac	Purebred Friesian	11½	25-7-44	22-2-45	Empty	Mastitis infection	Not milked in 1944/45 season Rearing calf	30 gm. thyroprotein daily over 14 days
Sophie	Jersey X Ayrshire Jersey	5½	28-1-45	22-2-45	Pregnant	Empty	Stilbestrol injected 5/6/44-19/9/44	30 gm. thyroprotein daily over 14 days
Nora	Purebred Friesian	11	6-8-45	25-7-46	Empty	Old age Mastitis	-	10 gm. thyroprotein Daily over 56 days
Ulex	Purebred Friesian	4	-	25-7-46	Empty	Sterile	-	10 gm. thyroprotein daily over 56 days

TABLE X (Cont'd.)

Name	Breed	Age at killing (years)	Last drying off date	Date of killing	Pregnancy status when killed	Reason for culling	Notes	Treatment in present experiment
Upstart	Grade Jersey	4	14-4-45	31-5-45	Empty	Sterile	Stilbestrol injected 5/6/44-19/9/44	Thyroprotein fed 24/11/44-18/12/44 7/1/45 - 27/1/45 24/2/45 - 16/3/45
Raupo	Grade Jersey	7	5-5-45	31-5-45	Empty	Empty	Stilbestrol injected 5/6/44-19/9/44	Thyroprotein fed 24/11/44-18/12/44 7/1/45 - 27/1/45 24/2/45 - 16/3/45
Tinder	Purebred Jersey	4	13-1-45	22-2-45	Pregnant	Empty	Stilbestrol injected 5/6/44-19/9/44	Thyroprotein fed 24/11/44-9/12/44
Quire	Grade Jersey	8	14-4-45	31-5-45	Empty	Empty	Stilbestrol injected 5/6/44-19/9/44	Thyroprotein fed 24/11/44-18/12/44 7/1/45 - 27/1/45 24/2/45 - 16/3/45
Ribbie	Purebred Friesian	7	5-5-45	31-5-45	Empty	Empty	Stilbestrol injected 14/10/43 27/12/43	Thyroprotein fed at intervals during lactation see Fig. II

the tissue from the middle of the right thyroid lobe.

The technique used for the histological examination of samples of the thyroids was as follows. The tissue samples were fixed in Bouin's picroformol immediately after being taken. After a period of fixation, they were embedded using the paraffin infiltration method. Cedarwood oil was the clearing agent employed. Prior to sectioning, the samples were soaked in lithium carbonate to soften the tissue. The sections were cut at 6 μ, stained with Mann's haematein and eosin, and mounted in green euparal.

Measurement of the Height of the Epithelial Cells

In histological studies of the thyroid gland by Rawson and Starr (1938), Rawson and Salter (1940, and Hertz and Roberts (1941), the degree of hyperplasia of the thyroid epithelium was measured by the height of the epithelial cells of the acini. The method involved the measuring of cells of average size of either 100 or 200 successive distinct acini. This method was criticised by Turner and Turner (1945) in that a possible error was introduced in determining by eye alone what cell or cells were of average height. To overcome this error, the latter workers used a photographic-planimetric method by which acinar cell area and colloid area were determined, and cell height was derived from these data.

Cognizance of the criticism of Turner and Turner was taken in the project here recorded, but as the equipment for such a photographic-planimetric method was not available, an alternative technique was developed to overcome the difficulty of deciding an average cell height by eye observation alone.

The first step was to calculate the number of cells required to provide a sample with a mean acinar cell height reasonably representative of that of all the acinar cells in the gland,

Samples of glands from two normal cows and one cow treated with thiourea and one cow treated with thyroprotein, were sectioned. Ten acini from each gland were randomly selected. The height of each of the epithelial cells in each of these ten acini was then measured, using an ocular micrometer calibrated from a stage micrometer. One division or unit of the ocular micrometer was equal to 1·25 μ. For convenience in the development of the sampling method, the measurements of acinar cell height were retained in units of the ocular micrometer, conversion to terms of microns being left until the final stages.

The observations on the height of the epithelial cells of the above four glands are recorded in Appendices VI to IX.

The mean acinar height for each of the ten acini of each gland is shown in Table XI.

An analysis of variance was applied to the mean acinar cell heights of the ten follicles of each of the glands and a standard deviation derived from the "within follicles" term of this analysis. These analyses are shown in Table XII.

With this knowledge of the mean cell height of ten randomly selected follicles of each of the four glands, and the standard deviation of the cell heights of the follicles within each gland, the size of the sample necessary to provide an accurate representation of the average cell height of each acinus could be

TABLE XI

Mean Acinar Cell Heights of Glands Used as the Basis for Sampling Technique

Name	Treatment	Mean Acinar Cell Height (in units) 1 unit = 1.25 μ										Mean for Gland
		Follicles										
		1	2	3	4	5	6	7	8	9	10	
Baronia	Normal	7.7	8.7	8.9	9.4	8.9	8.7	9.5	9.0	9.8	7.4	8.7
Qualify	Normal	7.7	9.9	10.0	9.4	9.6	8.9	10.2	9.0	10.5	7.9	9.5
Unmask	Thiourea	27.9	21.4	25.8	23.5	21.7	27.1	23.0	25.2	18.8	21.0	23.9
Quiver	Thyroprotein	5.0	3.9	4.7	5.1	4.7	5.5	5.1	5.2	4.6	5.1	4.9

TABLE XII

Analyses of Variance of Acinar Cell Height

Animal	Source	Degrees of Freedon	Sum of Squares	Mean Square	Standard Deviation
Normal cow "Baronia "	Total	1211	2743		
	Between Follicles	9	1055.31	117.26	
	Within Follicles	1202	687.69	0.5721	0.76
Normal cow "Qualify"	Total	995	3563		
	Between Follicles	9	2496	277.33	
	Within Follicles	986	1067	1.082	1.04
Cow fed thiourea "Unmask"	Total	840	21403		
	Between Follicles	9	8788	976.44	
	Within Follicles	831	12615	15.18	3.90
Cow fed Thyroprotein "Quiver"	Total	1030	1539		
	Between Follicles	9	160	17.78	
	Within Follicles	1021	1379	1.351	1.16

calculated.

The formula used was

$$n = \frac{t^2 s^2}{(\bar{x} - m)^2}$$

where n = the required number of observations; $\bar{X}$ = the mean cell height, and s = the standard deviation in cell height, of the sample of cells measured; m = population parameter cell height of the whole of epithelial cells within the gland; and t = 3, this value being arbitrarily chosen as being larger than the 1% level of the likely sample size (Snedecor, 1940 p.390).

The pooled standard deviation of the height of the acinar cells in the two normal glands is 0.9 units, and the average of the mean cell heights of those cells measured is 9.1 units. It is reasonable to assume, then, that the population parameter is about 10.1 units, as the best available measure of the mean acinar cell height of all the epithelial cells in the gland is the mean cell height of the sample of cells measured. On applying the above formula

$$n = \frac{(3)^2 \times (0.90)^2}{1} = 7.29 \text{ or } 8 \text{ observations}$$

Thus a sample of eight cells will be sufficient to show significance at the 1% level of a difference of 1 unit in mean acinar cell height between follicles. A table showing the difference in mean acinar cell height between follicles required to give significance at the 1% level for a varying number of observations in normal glands is shown below. The data shown in this table is presented in graph form in Figure IV.

In the thyroid gland of the cow treated with thiourea,

TABLE XIII

Difference in Mean Acinar Height Between Follicles Required to Give Significance at the 1% Level for a Varying Number of Observations in Normal Glands

Number of Observations	Required Difference in Means	
	Units	Microns
5	1.46	4
6	1.21	3
7	1.04	3
8	0.91	2
9	0.81	2
10	0.73	2
11	0.66	2
12	0.60	2
15	0.49	1
20	0.36	1
25	0.29	1
30	0.24	1

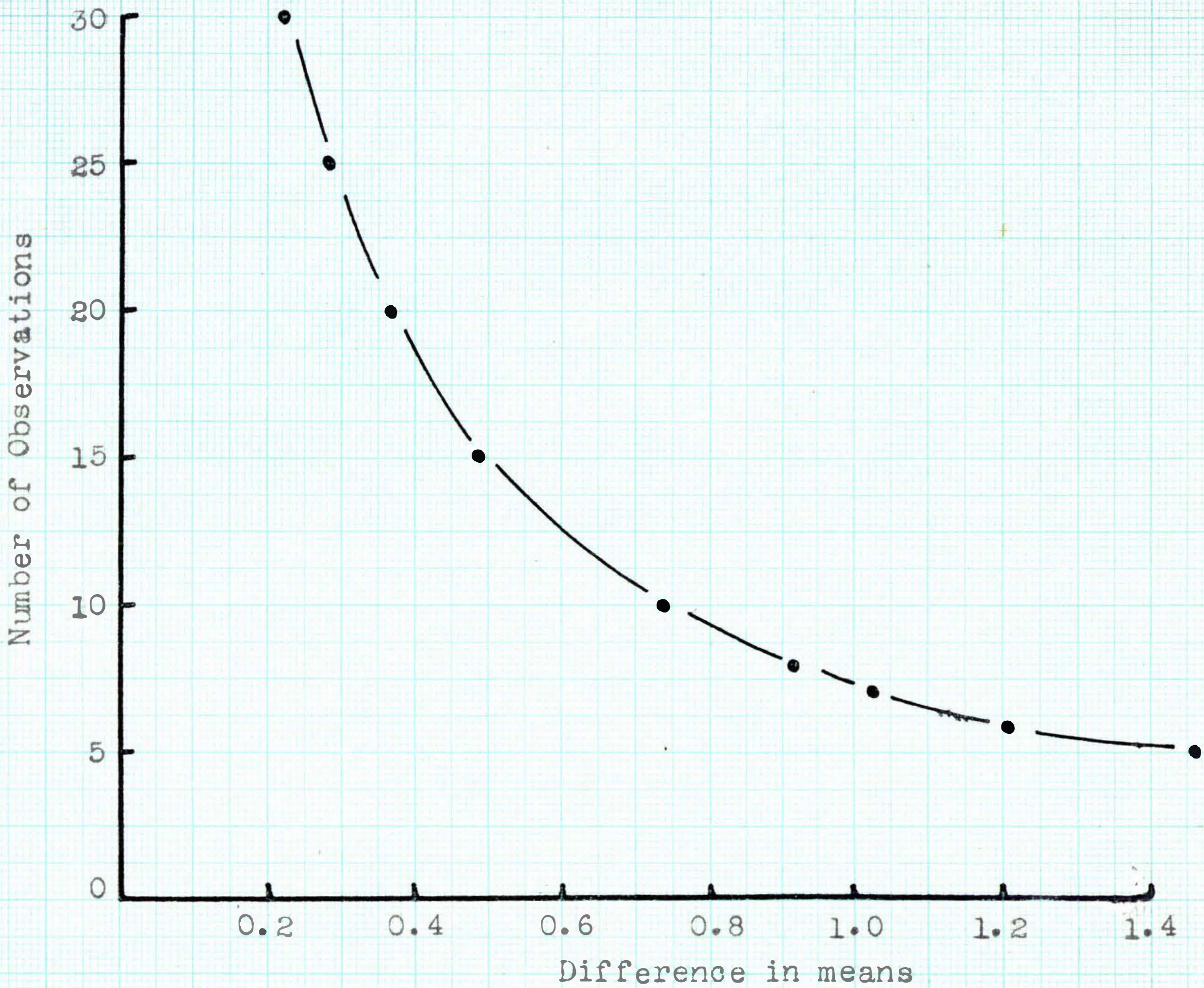


FIGURE IV

Number of Observations of Cell Height Required to Give a Difference in Means Significant at the 1% level

The variation in cell height was much greater than than found in normal glands. Horeover, the mean cell height of the ten follicles (23.9 units) was greater than that obtained for the two normal glands (8.7 and 9.5 units respectively). For the acinar cell heights of this gland from the thiourea-treated cow, a standard deviation of 3.90 units was found. In this case the number of observations necessary to show significance at the 1% level of a difference in mean acinar cell height between follicles of 7 units is

$$n = \frac{(3)^2 \times (3.90)^2}{7} = 19.54 \text{ or } 20 \text{ observations}$$

A table for such stimulated glands, analagous to that given above for normal glands, is as follows:-

TABLE XIV

Difference in Mean Acinar Cell Height Between Follicles Required to Give Significance at the 1% Level for a Varying Number of Observations in Stimulated Glands

Number of Observations	Required Difference in Means(Units)	
	Units	Microns
9	15	38
14	10	25
15	9	23
14	8	20
20	7	18
23	6	15
27	5	13

As the difference in means between follicles for such depressed glands as those from cows treated with thyroprotein is about 1.5 units, as compared with about 1 unit for normal glands, the table compiled for the latter glands may be utilised for depressed thyroids.

In order to determine the number of follicles that require to be measured from each gland to provide a representative sample, an analysis of variance covering all four glands was carried out in order to provide an "over-all" standard deviation in cell height.

TABLE XV

Analysis of Variance of Cell Height Between Glands from Two Normal Cows, from One Fed Thiourea, and From One Fed Thyroprotein

Source	Degrees of freedom	Sum of squares	Mean square
Total	4079	215,354	
Between glands	3	186,104	62035.00
Within glands	4076	29,250	7.176

Standard Deviation = 2.68

Applying the formula to this data,

$$n = \frac{(3.0)^2 \times (2.68)^2}{6} = 9.23\text{, or 10 observations}$$

Hence, by measuring cells from ten follicles, a difference of 6 units in mean values of glands should be significant at the 1% level for a stimulated gland. Because there is less variation in cell height within normal and depressed glands, and because the mean cell height of these glands is smaller than that in a thiourea-treated gland, in these cases $\bar{x} - m = 3$; thus for such glands a difference of 3 units in mean values of glands should be significant at the 1% level when cells from the follicles are measured.

From the above calculations it was thus found that to obtain a sample of cells with a mean acinar cell height reasonably representative of that of all of acinar cells in the thyroid gland, eight randomly selected cells from each of ten randomly selected follicles were all that were needed to be measured in a normal or depressed thyroid gland. (For the sake of convenience in calculation, ten cells from each of ten follicles were measured in these types of glands in this thesis). In a stimulated gland it was necessary to measure twenty cells from each of ten follicles. This sampling method was used in the histological examination of the thyroid glands in this thesis.

Effects of Thyroprotein

(a) General Effects. The trends in body weight and in pulse rate, where available, of the animals fed thyroprotein are shown in Figure V. In the case of the cows on the short-term feeding periods it will be seen that there was a marked increase in pulse rate accompanied by a gradual decline in body weight as the feeding period continued. There was no marked effect on the body weight of the two animals fed 10 gm. thyroprotein daily over 56 days, and the pulse rate, after an initial rise, declined again to only a slightly abnormal level and remained there for the remainder of the feeding period.

(b) Effects on Acinar Cell Height and Thyroid Gland Weight

(i) Thyroids of Animals Killed at the End of the Feeding Period. To study the effect of thyroprotein on the thyroid gland of cows being fed thyroprotein, eight cows were slaughtered the day following the conclusion of their respective feeding periods. The thyroid glands were weighed, and then examined histologically, and the mean acinar cell height for each gland measured. These results are shown in Table XVI, together

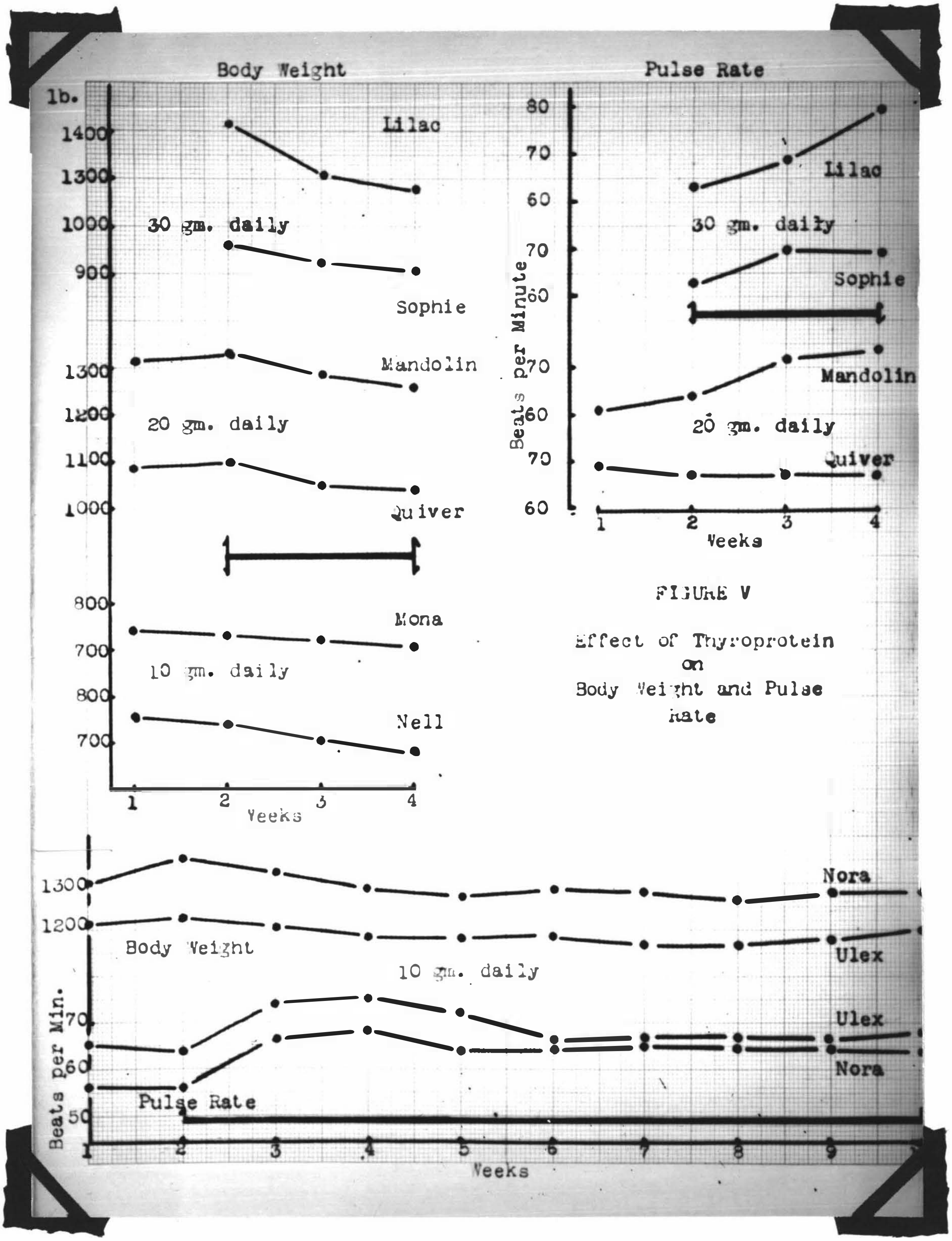


FIGURE V

Effect of Thyroprotein on Body Weight and Pulse Rate with the mean thyroid weight and mean acinar cell height of thyroids of normal cows of different breeds. Full data on the measurements of acinar cell height are given for normal glands in Appendicces X to XXII , and for glands from cows killed at the end of the thyroprotein feeding period in Appendices XXIII to XXIX.

TABLE XVI

Mean Acinar Cell Height and Gland Weight of Thyroids

Cow	Breed	Treatment	Mean acinar cell height (microns)	Mean gland weight (gm.)
	Jerseys	Normal	8·5	29.5
	Friesians	Normal	7·5	42.0
	Ayrshires	Normal	7·5	34.2
Mona	Jersey)	10 gm. daily	5·0	38.2
Nell	Jersey)	over 14 days	6·5	39.5
Quiver	Jersey)	20 gm. daily	6·0	48.0
Mandolin	Friesian)	over 14 days	4·5	36.0
Sophie	Jersey)	30 gm. daily	5·0	42.0
Lilac	Friesian)	over 14 days	4·5	17.0
Nora	Friesian)	10 gm. daily	[illegible]	45.4
Ulex	Friesian)	over 56 days	[illegible]	87.1

There were insufficient numbers of animals in each treatment to establish significant differences in acinar cell height between the various treatments. When, however, the glands from cows treated with thyroprotein are compared with those from normal animals, a highly significant difference was found between the mean acinar cell heights of each group (t=7.403), the acinar cellshof the thyroprotein-treated thyroids being abnormally low.

The effect of thyroprotein on the weight of the thyroid gland was analysed by testing the significance of the difference in mean thyroid weight between glands from the treated Jersey cows and the treated Friesian cows, and the mean thyroid weight found for the two respective breeds. The difference was non-significant both in respect to glands of the Jerseys (t=0.202) and the Friesians (t=1.418).

(ii) Thyroids of Animals Killed at Least 76 Days After End of Feeding Period. In view of the marked depression in acinar cell height found in the thyroids from cows slaughtered the day after the end of a thyroprotein feeding period, it was of interest to know whether such depressed glands would subsequently return to a normal condition, if no further thyroprotein were fed. To this end, the thyroid glands of five cows which had been on a thyroprotein feeding experiment concerned with a project outside the scope of this thesis, were also examined histologically, and the mean acinar cell height measured. Details of these animals have been given in Table X. The mean acinar cell heights and thyroid gland weights are shown in Table XVII, and full data from the histological examination are presented in Appendices XXX to XXXIV

TABLE XVII

Mean Acinar Cell Height and Gland Weight of Thyroids

Cow	Breed	Mean Acinar Cell Height (microns)	Thyroid Gland Weight (gm.)
Robbie	Friesian	11.0	51.5
Raupo	Jersey	10.5	24.0
Upstart	Jersey	10.0	33.0
Tinder	Jersey	13.5	18.0
Quire	Jersey	6.0	43.0

A test of significance showed that the mean acinar cell height of these glands did not differ significantly from that of normal glands (t=1.570). It is thus presumed that the experimental glands had returned to a normal condition at the time of slaughter, 76 days after the end of the thyroprotein feeding period.

REVIEW OF LITERATURE ON GOITROGENIC SUBSTANCES

Nature of Goitrogenic Substances

For many years the existence has been know of substances, both naturally-occurring and artificial, which are capable of inducing enlargement of the thyroid gland on being administered to animals and until recently it has been believed that the goitrogenic action of such substances was naturalised by the addition of iodine to the diet.

In 1941, MacKenzie, MacKenzie and McCollum reported that sulphaguanidine, used to combat intestinal infection, produced a remarkable enlargement of the thyroid gland in the rat. Histologically, the thyroids showed marked hyperplasia, the epithelium was distinctly columnar and was so increased and invaginated as nearly to extinguish the lumina of the follicles. MacKenzie and MacKenzie (1943) showed that a similar goitrogenic activity could be obtained with a series of sulphonamides and thioureas. The thyroid hypertrophy, which was accompanied by a fall in basal metabolic rate, was not prevented by the administration of iodine, but was inhibited by the injection of thyroxine. These findings were quickly confirmed (Astwood et al., 1943; Astwood, 1943a; Dempsey and Astwood, 1944), and similar results with substituted thioureas (Astwood et al. 1943; Richter and Clisby, 1942; Kennedy, 1942) and natural goitrogens (Griesbach and Purves, 1943) reported.

The thyroid hyperplasia induced by these goitrogens was accompanied by signs of increased activity of the anterior pituitary gland, but the thyroid hyperplasia could not be obtained in hypophysectomised animals (Griesbach and Purves, 1943). It was then thought that these substances depressed thyroid hormone production, and that the thyroid hyperplasia was caused by increased pituitary activity evoked by the reduced thyroid hormone output (MacKenzie and MacKenzie, 1943; Astwood et al., 1943). At about the same time it was observed that the prolonged administration of potassium thiocyanate to humans could induce the appearance of thyroid goitres, associated with a fall in basal metabolic rate, though the development of this type of goitre could be prevented by the administration of dietary iodine (Rawson, Hertz and Means, 1942).

Basing his therapy on the assumption that thiourea prevented the production of thyroid hormone, Astwood (1943b) successfully treated clinical hyperthyroidism by the daily administration of thiourea and showed that 2-thiouracil was also effective. The possibilities of this new treatment were soon widely explored (Rawson et al., 1944a; Paschkis et al., 1944; Astwood, 1944a; McGavick at al., 1944; Astwood, 1944a) and it was also shown that the administration of thiourea and thiouracil to experimental animals had effects similar to those of thyroidectomy with respect to growth (Hughes, 1944; Williams et al., 1944), organ morphology (Leblond and Hoff, 1944), thyrotrophin-induced metamorphosis of tadpoles (Hughes and Astwood, 1944), development of fish (Goldsmith at al., 1944a), and pigmentation of bird plumage (Juhn, 1944). Thiouracil, as mentioned previously, has been used to estimate the amount of thyroxine secreted under various conditions (Astwood et al., 1943; Astwood, 1943a; Dempsey and Astwood, 1944).

These results support the view that thioureas and thiouracil inhibit the formation of the thyroid gland hormone, but do not interfere with the action of the hormone once it has been liberated into the bloodstream.

Mode of Action of Goitrogenic Substances

It has been shown by Astwood and Bissell (1944) that the daily administration of thiouracil to young rats for eight days reduces the iodine content of the thyroid gland to a very low level although the weight of the gland may be increased nearly threefold. If daily administration of thyroxine is then commenced, with the continuation of the thiouracil treatment, the iodine content of the gland remains low but the follicles fill with densely staining colloid material (Astwood and Bissell, 1944; Dempsey, 1944). A similar result is obtained by hypophysectomy during thiouracil administration (Astwood and Bissell, 1944).

It appears that under these conditions the secreted colloid material contains little or no thyroxine, due to the inhibition by thiouracil of thyroxine formation.

On the injection of radio-active iodine into rats previously made goitrous by the administration of thiouracil, the power of the thyroid gland to collect the administered iodine may be only 10 to 20% of normal (Rawson et al., 1944b, 1944c, 1944d; Franklin et al., 1944a; Baumann, Metzger and Marine, 1944; Goldsmith et al., 1944b) and the formation of di-iodotyrosine and of thyroxine is also inhibited (Franklin et al., 1944a). The capacity of thiocyanate-induced goitres to collect administered radio-active iodine may be abnormally high (Rawson et al., 1944d) - a finding which is, significant in view of the fact that thiocyanate is an iodine-inhibited goitrogen.

Franklin et al., (1944b), working with surviving slices of thyroid tissue, found that the addition of thiouracil, of thiourea or of potassium thiocyanate reduced the ability of the tissue to convert added radio-active iodine into di-iodotyrosine and thyroxine.

Young (1944) has suggested that

> "The simplest hypothesis regarding the special function of the thyroid is that this gland possesses the special ability to collect iodine from circulation. Since free iodine is presumably the iodinating agent in the formation of di-iodotyrosine from tyrosine, and since iodide ions constituted the form in which this element is collected from the bloodstream, it seems probable that the first process which the collected iodide ions undergo is enzymic oxidation to free iodine. Inhibition of this process might not only inhibit the formation of di-iodotyrosine and therefore that of thyroxine but also depress the power of the gland to collect more iodide".

Werner (1912) observed that free iodine can oxidise thiourea to formamidine disulphide, being itself reduced to iodide ions in the process, and Campbell et al. (1944) have suggested that this may be a mechanism whereby thiourea might interfere with the synthesis of the thyroid hormone.

An alternative possibility is that thiourea and other similar goitrogens inhibit the action of an enzyme which catalyses the formation of iodine from iodide in the thyroid gland. This aspect of the problem has been investigated by Dempsey (1944) and Dempsey and Astwood (1944), and some evidence to support the theory has been found.

The present state of knowledge may be summed up by the statement that thiourea and thiouracil interfere with the formation of iodine from iodide ions, either by reducing any iodine formed back to iodide ions, or by poisoning the enzyme system catalysing the oxidation of iodide ions to iodine. It is still possible that these goitrogens may interfere in some further way with the formation of thyroxine in the body.

Toxic Effect of Goitrogenic Substances

The frequent occurrence of toxic effects of thiourea may limit its use for practical experimentation. The toxic effects of thiourea are usually manifested by digestive disturbances and if treatment is continued, emaciation and death may follow. Astwood (1943b) and Hinsworth (1944) noted that sulphonamides, thiourea and thiouracil may cause agranulocytosis in humans treated with them. Williams et al. (1946) reported that in humans thiouracil has in some cases caused toxic reactions. In the treatment of 247 patients, toxic reactions to the drug were encountered in 36, or 14.5% Thiourea was found by Schultze and Turner (1945) to be much more toxic than thiouracil at similar dosage levels for the chicken, goat and lactating cow.

Mixner, Reineke and Turner (1944) reported that thiourea was toxic to young chicks when administered at 0.6 to

0.8% levels in the drinking water. Schultze and Turner (1945) found that dosages of 0.15% thiourea or above, in the feed were toxic to chicks. The same workers found that a dosage of thiourea of 2.0 grams per day per 100 lbs. body weight, given orally was toxic to lambs, and that dosages of 0.75 grams per day given orally to goats of 50 to 100 lb. body weight were definitely toxic when given over a period of 30 days. No toxic effects of thiourea were noted in 150 lbs. calves when given in amounts as large as 3 grams per day orally for 30 days, but in cows even 5 grams thiourea per day per 1000 lb. liveweight was toxic as indicated by the fact that cows so treated went off feed and digestive disturbances were induced. Administration of 12 grams thiouracil per day to lactating cows caused no toxic effects.

Some evidence for the development of resistance to thiourea toxicity has been reported by Griesbach, Kennedy and Purves (1944) in rats, and by Schultze and Turner (1945) in lactating goats. In both cases the animals had previously survived a first dose of thiourea.

That the toxic effects of thiourea are independent of its goitrogenic effects is evident from the work of Schultze and Turner (1945) since goats receiving thiourea in addition to a large dosage of thyroxine exhibited toxicity symptoms as readily as those receiving thiourea alone.

Griesbach, Kennedy and Purves (1944) found that potassium iodide acted as a protective agent against thiourea toxicity in rats when given for a few days prior to thiourea treatment.

EXPERIMENTAL

Effedts of Thiourea on Dairy Cows

(a) Methods. The experiments described in this thesis were undertaken to study the effect of thiourea on the thyroid gland of the dairy cow.

Details of the animals used and dosages given are shown in Table XIX.

The thiourea was dissolved in warm water and given as a drench to dry dairy cows. During the experimental period the animals were weighed and their pulse rates determined where possible, onthree consecutive days weekly. On the day after the conclusion of the feeding period the animals were slaughtered. The dissecting out of the thyroid, weighing of the gland, sampling for histological examination and histological methods were carried out as outlined in the earlier section concerning animals fed thyroprotein.

The trends in body weight and in pulse rate, where available are shown in Figure VI.

(b) General Effects. The trends in body weight and pulse rate where available of the cows fed thiourea, are shown in Figure VI. It will be observed that during the feeding period there was a tendency for the pulse rate to decline, this being accompanied by a slight decline in body weight.

The decline in body weight was probably due to toxic effects of the thiourea, for the animals were observed to lose their appetites to some degree within a few days of commencing the thiourea feeding period. The material obviously was dis-

TABLE XIX

Details of Cows Fed Thiourea

Name	Breed	Age at killing (years)	Last drying off date	Date of killing	Pregnancy status when killed	Reason for culling	Notes	Treatment in present experimt
Quack	Purebred Jersey	8	3-6-44	1-3-45	Empty	Empty	Stilbestrol injected 5/6/44 - 19/9/44	10 gm. thiourea daily over 7 days followed by 15gm daily over 14 days
Union	Purebred Jersey	5	9-3-46	11-4-46	Empty	Empty. Low production	-	10 gm. thiourea daily over 7 days followed by 15gm daily over 14 days
Upright	Grade Jersey	5	16-3-46	11-4-46	Empty	Empty. Low production	-	25 gm. thiourea daily over 21 days
Unmask	Jersey X Ayrshire-Jersey	5	9-2-46	11-4-46	Empty	Empty. Low production	-	25 gm. thiourea daily over 21 days

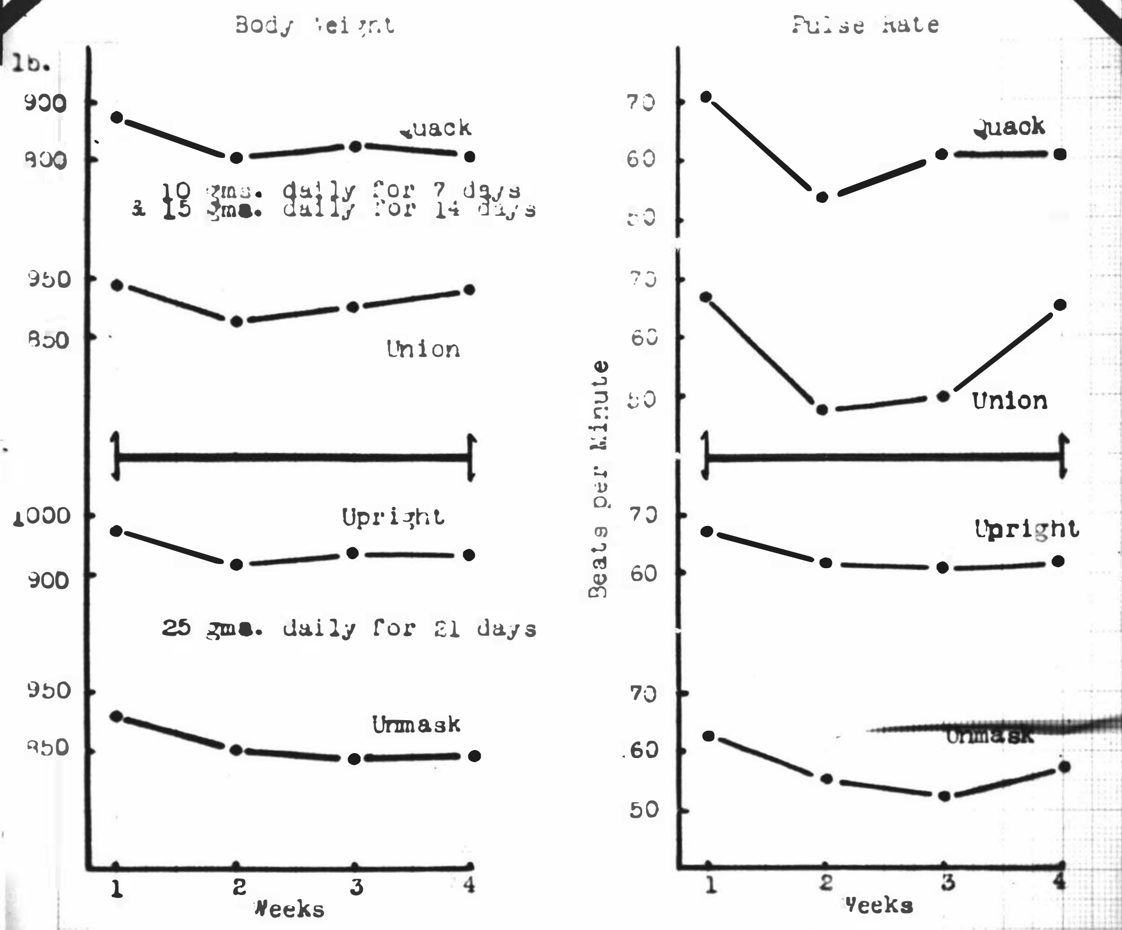


FIGURE VI

Effect of Thiourea on Body Weight and Pulse Rate tasteful to the animals as manifested in refractory behaviour at dosing, and coughing and tossing of the head after dosing. On the day prior to killing, one cow, Union, on a dose level of 10 gm. thiourea per day over 7 days followed by 15 gm. per day over 14 days, showed excitability, as reflected in a marked increase in pulse rate, twitching in muscles of the limbs, and flickering of the eye-lids.

(c) Effect on Acinar Cell Height and Thyroid Weight. To study the effect of thiourea on the acinar cell height and thyroid gland weight of dairy cattle, four Jersey cows were fed thiourea and slaughtered on the day after the end of the feeding period. The mean acinar cell height and gland weight of the thyroids from these animals are shown in Table XVIII. Complete data on the height of the epithelial cells of these thyroids are given in Appendices XXXVI to XXXVIII

TABLE XVIII

Mean Acinar Cell Heights and Thyroid Weights

Cow	Breed	Treatment	Mean acinar cell height (microns)	Thyroid gland weight (gm.)
	Jerseys	Normal	8.5	29.5
Quack	Jersey	10 gm. thiourea daily over 7 days followed by 15 gm. daily over 14 days	19.0	39.0
Union	Jersey		24.0	33.8
Unmask	Jersey	25 gm. thiourea daily over 21 days	30.0	46.8
Upright	Jersey		29.8	37.4

A test of significance was applied to the difference in mean acinar cell height between the glands from the thiourea-treated animals as a group and the glands from normal cows. The mean acinar cell height of the glands from the thiourea-treated cows was found to be highly significantly greater than that of normal cows (t=9.070). The number of cows in each group was too small to allow of statistical analysis of differences in acinar cell height between treatments.

The mean weight of the thyroid glands of thiourea-treated animals was found to be significantly greater than that of normal Jersey cows (t=2.190)

DISCUSSION

Although the data obtained on the weights of the thyroid glands of various classes of cattle are as yet limited, they provide an indication of the weights of the thyroid glands of cattle in the Manawatu district. The data are already proving valuable in that glands from abnormal animals may be compared with them to reveal possible abnormalities in thyroid weight. In this way the data are providing a basis for study of the etiology of abnormal conditions manifested in thyroid dysfunction. A case in point is that among calves born from dams that have been running on Manawatu sandy loam, stony phase for some time, animals have been found with thyroid glands which, on comparison with the glands collected from seventy-five calves from dams running on different soil types within the same district, are unusually large. Attention is thus directed towards factors affecting thyroid function of animals on this soil type.

The collected data on the thyroid weight of dairy cows has been of use in the experiments with thyroprotein and thiourea recorded in this thesis in that the average thyroid weight of a reasonably large number of normal dairy cows was thus made available for comparison with the thyroids from the experimental animals

With the accumulation of further thyroid weight data, particularly in respect to thyroids of dairy cows, both in full milk and dry, it will be possible to study the variation in thyroid weight due to such factors as season of the year, lactation and district, and to further study the influence of age of the animal on the thyroid weight. Glands from dairy cows in full milk and from dairy heifers of 1 to 2 years of age are, however, very difficult to obtain, for such animals are rarely seen in the slaughter-houses. It is only over a period of years, therefore, that such data will become available.

In the analysis of the thyroid weight data according to breed, it was found that the differences between the mean thyroid weights of the three breeds of dairy cattle was statistically significant. These differences in mean thyroid weight were very similar to those obtained by Swett et al. (1937) who collected data from dairy cattle slaughtered in various parts of the United States. Moreover, the absolute mean thyroid weight obtained by Swett for cows of each of the three dairy breeds, Jersey, Friesian, and Ayrshire, were very similar to those obtained for cows of the same breeds in the Manawatu.

From the data presented by Krizenecky (1932), it appears that dairy cattle have larger and heavier thyroids than beef cattle, averaging 32.5 gm. as compared with 24.2 gm. for beef cattle. No data was given, however, showing variation due to breed. The analysis made in this manuscript confirms the conclusion of Krizenecky in that the mean thyroid weight of Friesian cows of about 1250 lb. body weight was shown to be significantly greater than the mean thyroid weight of Aberdeen Angus cows of approximately 1300 lb. body weight. Reece and Turner (1937) have shown that the thyrotrophin content of the pituitary gland of dairy cattle is higher than that of beef cattle, which may explain, in part the larger thyroid gland of dairy cattle. It appears, therefore, that the weight of the thyroid gland is dependent upon the physiological function of the animal, i.e. beef or milk production.

The finding of no significant difference in thyroid weight due to sex in the groups compared - male and female Jersey calves, castrated male and intact female Aberdeen Angus vealers, and cows and steers of the Aberdeen Angus breed - is in conformity with the results of Mixner, Reineke and Turner (1944) with chicks, and Goldsmith, Gordon and Charipper (1945) with rats.

The technique that was developed for measuring the mean acinar cell height of the thyroid glands proved to be satisfactory. That the number of acinar cells calculated to be necessary to establish significance at the 1% level of differences between the means of the postulated magnitude was sufficient, was proved by the highly significant diference in mean acinar cell height found between glands from normal cows and cows fed thyroprotein, and normal cows and cows fed thiourea, respectively.

The studies reported in this thesis on the effect of thyroprotein on the thyroid gland of the dairy cow showed that dose levels of thyroprotein even as low as 10 gm. per day resulted in an increase in pulse rate and a decrease in height of the thyroid epithelial cells. The animal was capable of assimilating sufficient extra nutrients to meet this increase in metabolism without loss of body weight, as shown in Figure V. This finding if it can be confirmed, will be useful in the future use of thyroprotein in experimental work.

The physiological process by which the thyroid acinar cell height is depressed on the feeding of thyroprotein involves the delicate mechanism regulating the thyrotrophic activity of the pituitary and the activity of the thyroid gland. The increased concentration of thyroxine in the bloodstream, as a result of thyroprotein administration, probably retards the output of thyrotrophin by the pituitary gland. This lowered output of thyrotrophic hormone results in a decrease in the activity of the thyroid gland, and hence causes a decrease in thyroid hormone output and in the height of the thyroid epithelial cells.

An interesting situation illustrated in this thesis was that the thyroid glands of cows fed thyroprotein and then managed in a normal manner until slaughter, 76 days after the end of the thyroprotein-feeding period, had returned to a normal condition during the post-feeding period. There was no significant difference between mean acinar cell heights of these animals (21 μ) and those of normal animals (19 μ). This was so, even in the case of Robbie, a cow that was thyroprotein fed at frequent intervals over 14 months (see Figure 11). An analogous situation has been described in cases where estrogens have been administered to non-pregnant dairy cows as a means of inducing lactation (Hammond and Day (1944), Folley and Malpress (1944), and Hollard and Campbell (1946). After the cessation of estrogen treatment, in many cases the estrous cycles returned to normal and pregnancy ensued.

The main object of feeding thiourea to dairy cows in the experiments recorded in this thesis was to determine a dose level that would have a definite effect on the thyroid gland without being too toxic to restrict its use in experimental work. It was found that a dose level of 10 gm. daily for 7 days followed by 15 gm. daily for 14 days caused a marked increase in thyroid acinar cell height. However, even at this dose level, the lowest used, one cow showed symptoms of acute toxicity, while others on the same dose level showed no such acute symptoms. It thus appears that the dose levels used were on the border-line of acute toxicity.

Future experiments may be planned along two lines. The effect should be determined of a lower dose rate of thiourea, say 5 gm. daily, with which the toxic effects of thiourea should not be shown. Secondly, other goitrogenic substances may be used without fear of toxic effects on the animal. One such compound is thiouracil. Supplies of this material were not available for use in the projects reported in this thesis, but it is anticipated that it will become available in sufficiently large quantities for use with dairy cows. Though not so potent in its goitrogenic properties as thiourea, thiouracil is not nearly as toxic as thiourea to humans (Williams, 1946) and smaller animals (Schultze and Turner (1946), and so it may be even more useful in thyroid studies with dairy cows than is thiourea.

With a dose level of thiourea or thiouracil established that is non-toxic yet goitrogenic in its effects on the dairy cow, it will be possible in conjunction with thyroxine to estimate the hormone output of the thyroid of high and low producing cows, and of lactating cows at all seasons of the year. Such knowledge would be of fundamental value in providing further information on the physiology of milk secretion.

SUMMARY

1. The general anatomy of the thyroid gland has been described and the anatomy of the thyroid gland of cattle was found to agree with the general description.

2. Thyroid glands were collected at the Palmerston North Municipal Abattoirs, Longburn Freezing Works, and Feilding Freezing Works from the following classes of cattle:- male and female Jersey calves; dairy cows of the Jersey, Ayrshire, and Friesian breeds; Jersey bulls; Aberdeen Angus vealers, heifers, cows and steers.

3. The mean thyroid weights of cows of the three dairy breeds were Jerseys, 29.5 gm.; Ayrshires, 34.2 gm.; and Friesians, 42.0 gm. The difference in mean thyroid weight between each of the three breeds was shown to be statistically significant.

4. Animals of each of the dairy breeds were shown to have a heavier thyroid weight per unit of body weight than the Aberdeen Angus cows.

5. No significant difference in thyroid weight due to sex was found between male and female Jersey calves, castrated male and intact female Aberdeen Angus vealers, and cows and steers of the Aberdeen Angus breed.

6. No significant increase in thyroid weight was found between Jersey cows of 3 years of age and those of 6 years of age or more. There was, however, a tendency towards increasing thyroid weight with advance in age.

7. The functioning of the thyroid gland has been described,

with particular reference to the chemistry of the thyroid hormone, the relation of iodine to the thyroid gland, and the regulation of thyroid activity.

8. The physiological activity of iodinated proteins has been discussed.

9. Experiments were undertaken to obtain some measure of the potency of the thyroprotein that was prepared at the Dairy Research Institute (N.Z.). It is estimated that this material was slightly less potent than the "Protamone" used by United States workers, but of approximately the same potency as the iodinated casein preparations used in investigations in England.

10. A method that conformed with statistical requirements was developed to select a sample of acinar cells with a mean cell height reasonably representative of all the acinar cells of a thyroid gland.

11. Using this method, the mean acinar cell heights of the thyroid glands of 13 normal dairy cows were estimated.

12. To study the effect of thyroprotein on the thyroid gland of the dairy cow, dosages ranging from 10 gm. to 30 gm. daily over 14 days were administered to eight dairy cows. These animals were slaughtered at the end of the thyroprotein-feeding period. The trends in body weight and pulse rate of these animals during the feeding period are presented.

13. The mean acinar cell heights of the thyroids of the thyroprotein-fed animals were measured. The overall mean acinar cell height of these thyroids (5μ) was shown to be significantly lower than that of the overall mean of the thyroids from normal dairy

cows (19 μ).

14. The difference in thyroid weight between cows fed thyroprotein and normal cows of the same breed was shown to be non-significant.

15. The mean acinar cell height of thyroids of five cows fed thyroprotein but not slaughtered until 76 days after the end of the feeding period was measured. There was shown to be no significant difference between the mean cell height of these glands and that of glands from normal dairy cows of the same breed.

16. The nature of goitrogenic substances, their mode of action, and their toxicity have been discussed.

17. To determine a dose rate of thiourea that would stimulate thyroid activity without being toxic to the animal, this material was fed to four dry dairy cows at dose levels ranging from 10 gm. daily over 7 days followed by 15 gm. daily over 14 days, to 25 gm. daily over 21 days. The animals were slaughtered at the end of the thiourea-feeding period.

18. The mean acinar cell height of thyroids from cows fed thiourea (26 μ) was shown to be highly significantly greater than that of normal cows (19 μ).

19. The mean thyroid weight of cows fed thiourea was significantly greater than that of normal dairy cows.

20. The effect of thyroprotein on the thyroid gland of dairy cows has been discussed.

21. The effect of thiourea in relation to its toxic reactions as a means of inducing hypothyroidism in dairy cows has been discussed.

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PLATES

PLATE 1.

THYROID GLAND OF A NORMAL DAIRY COW (X90).
SECTION OF THE THYROID GLAND OF "BARONIA".

(See p.25).

PLATE 11.

THYROID GLAND OF A DAIRY COW FED THYROPROTEIN (X90). SECTION OF THE THYROID GLAND OF "LILAC", FED 30gm. THYROPROTEIN DAILY OVER 14 days.

(See p.25).

PLATE 111.

THYROID GLAND OF A DAIRY COW FED THIOUREA.
SECTION OF THE THYROID GLAND OF "UNMASK", fed 25 gm. THIOUREA DAILY OVER 21 days.

(See p.48).

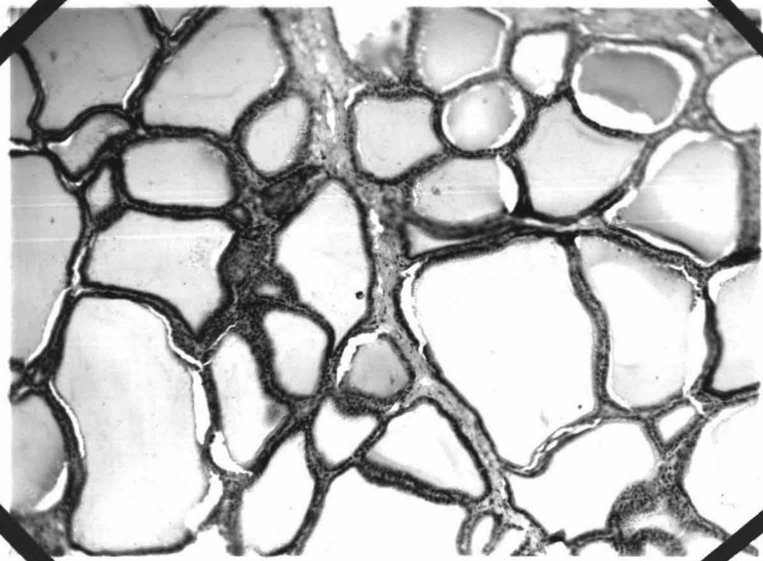

PLATE I

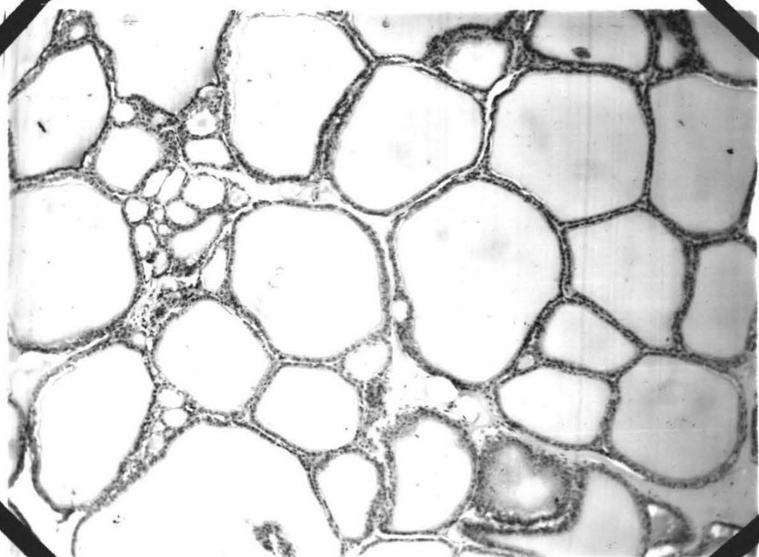

PLATE II

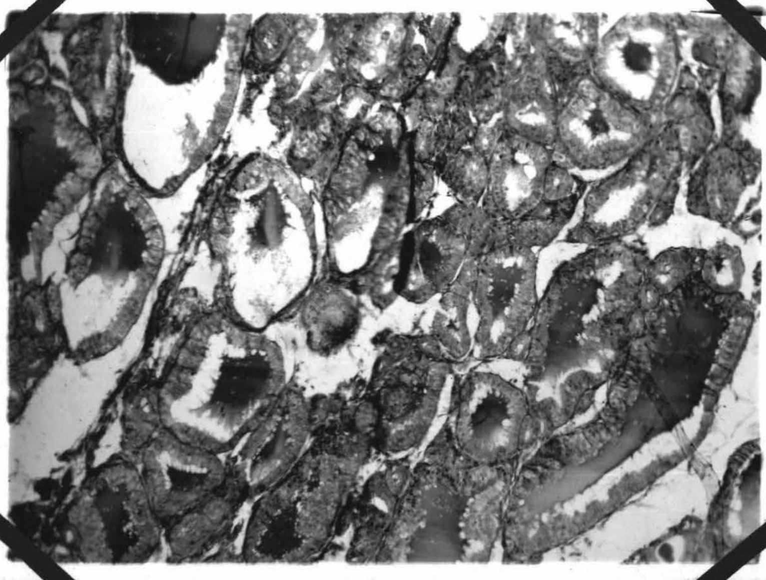

PLATE III

PLATE 1V.

THYROID GLAND OF NORMAL DAIRY COW (X375).
SECTION OF THE THYROID GLAND OF "BARONIA".

(See p.25).

PLATE V.

THYROID GLAND OF A DAIRY COW FED THYROPROTEIN (X375). SECTION OF THE THYROID GLAND OF "LILAC", FED 30 gm. THYROPROTEIN DAILY OVER 14 days.

(See p.25).

PLATE V1.

THYROID GLAND OF A DAIRY COW FED THIOUREA.
SECTION OF THE THYROID GLAND OF "UNMASK", FED 25 gm. THIOUREA DAILY OVER 21 Days.

(See p. 48).

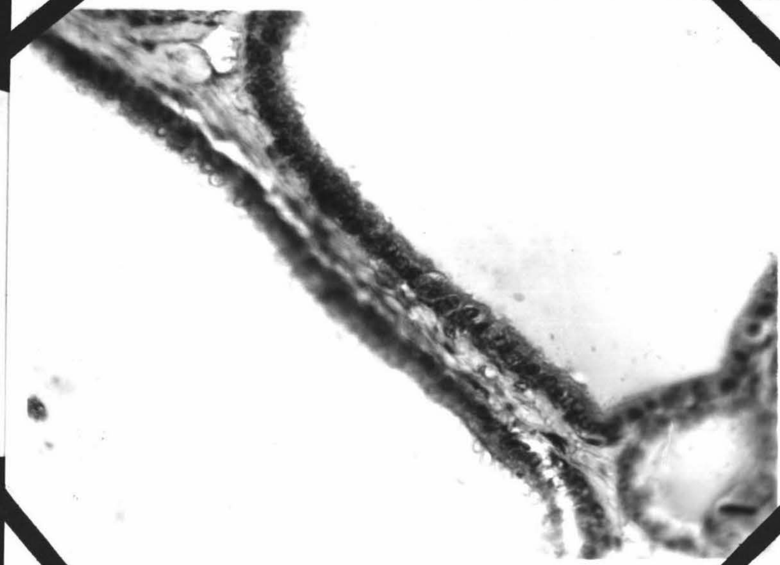

PLATE IV

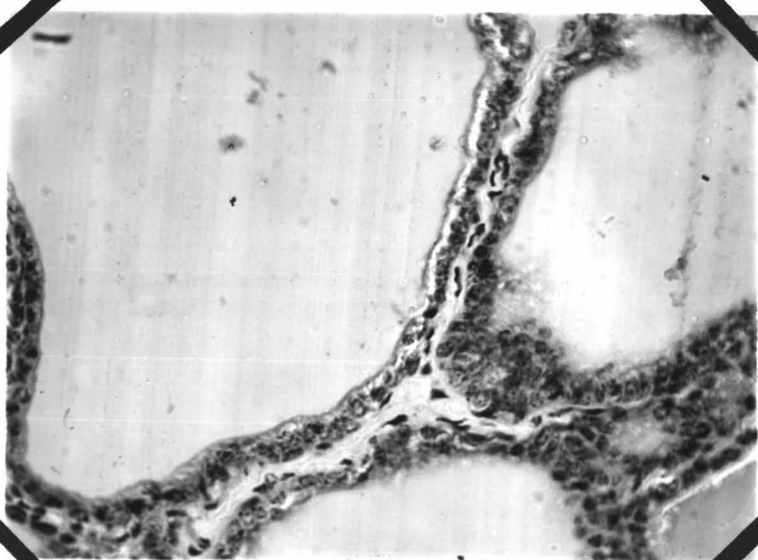

PLATE V

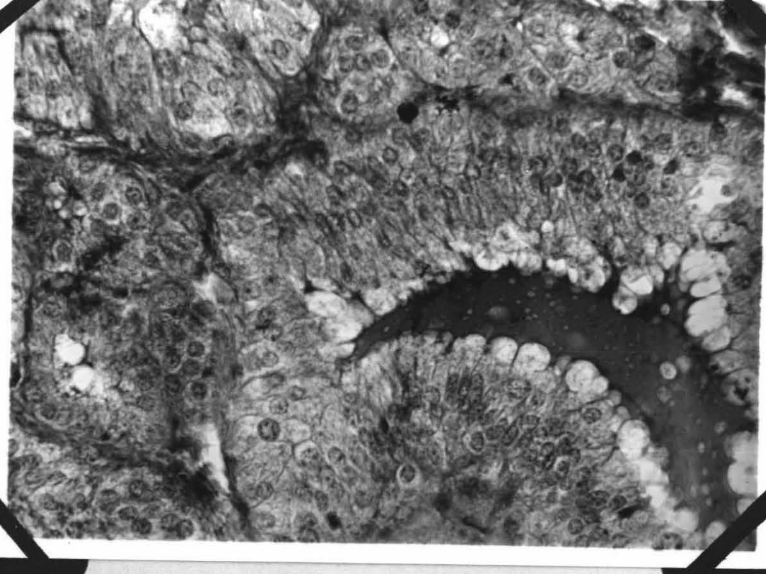

PLATE VI

APPENDIX I

Thyroid Weights of Dairy Cows

Jerseys				Friesians	Ayrshires
17.6 grams	15.3 grams	50.5 grams	20.5 grams	56.5 grams	32.5grams
22.1	20.0	25.7	27.9	31.2	23.8
26.3	30.0	38.0	26.5	25.1	37.7
28.0	18.2	16.6	23.9	32.1	26.1
27.4	17.5	29.8	24.2	55.7	32.8
25.6	19.5	44.3	19.3	42.0	37.5
29.2	24.0	48.7	20.6	56.0	42.0
34.7	31.0	33.9	32.5	37.7	32.3
33.3	33.0	20.4	41.6		38.5
27.1	49.0	25.2	31.4		39.1
29.1	43.0	27.0	44.7		25.5
19.1	30.0	36.0	29.8		32.8
47.5	37.0	37.2	38.6		44.7
37.4	30.0	27.5	23.8		27.7
40.6	28.9	47.3	27.0		37.5
11.9	20.7	27.7	28.0		34.0
27.2	38.2	42.5	28.8		31.5
23.7	30.5	35.3	36.5		39.0
8.5	22.3	19.0	22.7		30.5
	32.5	40.2	33.1		38.7
		30.9	31.0		
Total number of glands		=	81	8	20
Mean Weight (grams)		=	29.5	42.0	34.2

APPENDIX II

Thyroid Weights of Jersey Calves

Longburn Works				Massey College
Males		Females		Males
6.0 grams	8.2 grams	7.2 grams	5.8 grams	11.3 grams
5.6	7.1	6.2	8.5	8.6
4.9	5.0	8.3	5.2	9.7
5.2	9.0	6.9	8.6	9.9
4.9	5.0	4.8	5.5	10.6
5.5	6.2	3.8	13.2	10.1
7.5	7.4	5.2	8.5	8.0
7.9	7.0	10.9	7.6	
4.9	11.8	7.6	11.6	
7.6	5.7	7.6	10.9	
8.0	7.1	6.1	5.8	
5.2	7.2	8.5	8.8	
7.2	8.2	6.0	12.4	
6.4	7.6	13.5	8.0	
11.9	10.6	8.4	5.4	
16.0	8.4	8.1	4.5	
7.1		7.6	4.6	
		4.2	6.0	
		6.0	9.7	
		7.7	9.7	
		5.8	10.3	
Total number of glands				
33		42		7
Mean Weight (grams)				
7.4		7.6		9.7

APPENDIX III

Thyroid Weights of Steers

Kairanga District		Hunterville and Fordell Districts	
42.8 grams	24.5 grams	24.6 grams	33.0 grams
26.0	21.5	32.5	22.0
34.7	37.5	43.0	39.0
34.5	27.0	33.0	26.0
30.6	25.3	23.0	25.0
25.5	27.4	26.0	21.0
24.8	22.0	37.0	21.0
21.8	26.5	34.0	41.0
26.3	29.2	24.0	22.0
24.3	28.8	25.0	34.0
18.0	30.5	33.0	23.0
25.0	36.8	32.0	22.0
19.9	36.3	34.0	28.0
28.7	54.8	36.0	24.0
22.0	43.9	21.5	29.0
33.7		31.0	25.0
		32.0	23.0
		25.0	24.0
		30.0	22.0
		21.0	22.0
		26.0	
Total number of glands	31		41
Mean Weight (grams)	29.4		28.0

APPENDIX IV

Thyroid Weights of Vealers

Males		Females	
17.0 grams	17.0 grams	30.0 grams	16.5 grams
27.0	14.5	19.5	14.0
18.0	18.5	15.5	15.5
14.5	13.5	13.5	16.5
13.0	16.0		
17.0	16.5		
14.0	17.0		
14.0	14.5		
17.0			

	Males	Females
Total number of glands	17	8
Mean Weight (grams)	16.4	17.6

APPENDIX V

Thyroid Weights of Beef Heifers and Beef Cows

Beef Heifers		Beef Cows
January - February	Late March	January - March
33.3 grams	24.5 grams	30.2 grams
26.5	26.0	28.0
29.4	26.7	33.1
34.2	47.7	24.3
21.0	29.0	30.5
29.4	22.4	31.4
21.8	37.2	31.9
31.2	26.2	36.2
15.5	23.4	30.4
21.0	43.0	23.5
27.6	22.2	33.0
16.1	35.7	31.4
28.0	38.2	39.8
17.4	39.8	37.5
18.1	29.0	36.3
17.0	26.7	
23.2	22.6	
	26.5	
	41.8	
Total number of glands		
17	19	15
Mean Weight (grams)		
24.1	31.0	31.8

APPENDIX VI

Thyroid Acinar Cell Heights (in Units) of a Normal Dairy Cow "Baronia"
(1 unit = 2.5 μ)

Follicle number

1			2			3			4			5			6			7			8			9			10		
6	7	9	5	7	6	8	9	8	8	10	10	8	9	8	8	8	9	9	10	11	8	9	10	10	11	9	8	7	7
6	7	9	5	7	6	8	9	8	8	10	10	8	9	8	8	8	9	9	10	11	8	9	10	10	11	9	8	7	7
6	7	9	5	7	7	8	9	8	8	9	10	8	9	8	8	8	9	9	9	11	8	9	10	10	10	9	8	7	7
5	7	9	5	7	7	8	10	8	8	9	9	8	9	8	8	8	9	9	9	10	8	9	10	10	10	9	8	7	
5	7	9	5	7	7	8	10	8	8	9	9	8	9	8	8	8	10	9	9	10	8	9	10	10	10	9	8	7	
6	7	9	5	7	7	8	10	9	8	9	9	10	9	8	8	8	10	9	9	10	8	9	9	10	10	9	8	7	
6	7	9	5	7	7	8	10	9	8	9	9	10	9	8	8	8	10	9	9	10	8	9	9	10	10	9	8	7	
6	7	9	5	6	7	8	10	9	8	8	9	10	8	8	8	8	10	9	9	11	8	9	9	10	10	9	8	7	
6	8		5	6	7	8	10	9	8	8	9	10	9	8	8	8	10	9	9	11	8	9	9	10	10	9	8	7	
7	8		5	6	7	8	10	9	8	8	9	10	9	9	8	8	10	9	9	11	8	9	9	10	10	9	8	7	
7	8		6	6	7	8	10	9	8	8	9	10	9	9	8	8	10	9	9	11	8	9	9	10	9	9	8	7	
7	8		6	6	7	8	10	9	8	8	9	10	9	9	8	8		9	9	11	8	9	9	10	9	9	8	7	
6	8		6	6	7	8	10	9	8	8	9	10	9	9	8	8		9	9	11	8	9	9	10	9	10	8	7	
6	8		6	6	7	9	10	9	9	8	9	9	10	9	8	8		9	9	11	8	9	9	10	9	10	8	7	
6	8		6	6	8	9	10	9	9	8	9	9	10	9	8	8		9	9	10	8	9	9	10	9	10	9	7	
6	8		6	6	8	9	10	9	9	8	9	9	10	9	8	8		9	9	10	8	9	9	10	9	10	9	7	
6	8		6	6	8	9	11	9	9	8	9	9	10	9	9	8		9	9	10	9	9	9	10	9	10	10	7	
6	8		6	6	8	9	11	8	9	9	9	9	10	9	9	8		9	9	10	8	9	9	10	9	10	10	7	
7	8		6	7	8	10	11	8	9	9	10	9	10	9	9	9		9	9		8	9	9	10	9	11	10	7	
7	8		6	7	8	10	11	8	9	9	10	9	10		9	9		9	9		8	9	9	10	9	11	10	7	
8	8		6	7	8	10	11	8	9	9	10	9	10		9	9		9	9		8	10	10	10	9	11	10	7	
8	8		6	7	8	10	11	8	10	9	10	9	10		9	9		9	9		8	10	10	10	9	11	10	7	
8	8		6	7	8	10	11	9	10	9	10	9	10		9	9		9	9		8	10	9	10	9	11	10	7	
8	8		6	7		10	10	9	10	9	10	9	10		9	9		9	9		8	10	9	10	9	10	10	7	
8	8		6	7		10	10	9	10	9	10	9	10		9	9		9	9		8	10	10	10	10	9	10	7	
8	8		6	7		8	10	9	10	9	10	9	10		9	9		9	9		8	10	10	10	10	9	10	7	
8	8		7	7		8	10	9	10	9	10	9	10		9	10		9	9		8	10	9	10	10	9	10	7	
8	8		7	7		8	10	9	10	9	10	9	10		9	10		9	9		8	10	10	10	10	11	10	7	
7	8		7	7		8	10	8	10	9	10	9	10		9	10		8	9		8	10	10	10	10	11	10	7	
7	8		7	8		8	10	8	10	9	10	9	9		9	10		8	9		8	10	10	10	10	11	10	8	
7	8		7	8		8	10	8	10	9	10	8	9		9	10		8	9		8	10	9	10	10	11	10	8	
7	8		7	7		8	10	8	10	9	10	8	9		9	10		8	9		8	10	10	10	10	11	9	8	
7	9		7	7		8	10	8	10	9	10	8	9		9	10		8	10		8	10	10	10	10	10	9	8	
7	9		8	6		8	10	8	10	10	10	8	9		9	10		8	10		8	10	10	10	10	9	9	8	
7	9		8	6		9	10	8	10	10	10	8	9		9	10		8	10		8	10		10	10	9	9	8	
7	9		8	6		9	9	8	11	10	10	8	8		9	10		9	10		8	10		10	9	9	9	8	
7	9		8	6		9	9	8	11	10	10	8	8		9	10		9	10		8	10		10	9	11	8	8	
7	9		8	6		9	9	8	11	10	10	8	8		9	9		9	10		9	10		10	9	11	8	8	
7	9		8	6		9	9	8	11	10	10	8	8		9	9		9	10		9	10		10	9	11	8	8	
7	9		8	6		9	9	9	11	10	10	8	8		8	9		10	10		9	10		10	9	11	8	8	
7	9		8	6		9	9	9	11	10	10	8	8		8	9		10	10		9	10		10	9	10	8	8	
7	9		8	6		9	9	9	10	10	10	8	8		8	9		10	10		9	10		10	9	10	8	8	
7	9		8	6		9	9	9	10	10	10	9	8		8	9		10	11		9	10		10	9	9	8	8	
7	9		8	6		9	8	9	10	10	10	9	8		8	9		10	11		9	10		10	9	9	8	8	
7	9		8	6		9	8	9	10	10	10	9	8		8	9		10	11		9	10		10	9	9	9	8	
7	9																												

APPENDIX VI (Cont'd)

Follicle number

	1	2	3	4	5	6	7	8	9	10
	7 9 7 9	7 6 7 6	9 8 8 9 8 8 8 9 9 8 9 9 8 8 9 8 8 9 8 8 9 8 8 8 8 8 8 9 8 8 9 8 8 9 8 8 9 8 8 9 9 8	10 10 10 10 10 10 10 10	9 8 9 8	8 9 8 9	10 11 10 11	9 10 9 10	11 9 11 11 9 11 11 10 9 11 10 9	7 8 7 7
Sum	783	779	1567	1337	913	851	1189	1157	1424	565
No. of cells	102	117	177	142	103	98	125	128	145	75
Mean cell heights	7.7	6.7	8.9	9.4	8.9	8.7	9.5	9.0	9.8	7.5
Sum of squares	6044	5279	14001	12681	8153	7433	11385	10537	14050	4275

Mean of mean cell heights of follicles = 8.7 units

APPENDIX VII

Thyroid Acinar Cell Heights of a Normal Dairy Cow "Qualify"

Follicle Number

1	2	3	4	5	6	7	8	9	10
8 7 8	10 10 10 10	10 11 11 10	10 10 9 6	10 10 11 9	8 10 10 10	10 13 9 10	7 7 8 8	9 9 9 9	10 10 10 10
8 7	10 10 10 10	10 11 11 10	10 10 9 10	10 10 10 9	8 10 10 10	10 13 10 10	10 10 10 10	8 10 11 11	10 10 10 10
8 7	10 11 10 10	10 11 11 10	10 9 9 10	10 10 10 9	8 10 10	10 13 10 10	11 11 11 9	8 10 11 11	9 9 9 9
8 8	10 11 10 10	10 11 11 10	10 9 9 10	10 10 10 9	8 10	10 13 10 10	11 9 9 9	8 10 11 11	9 8 8 8
8 8	10 11 10 10	10 11 10 9	10 9 9 10	10 10 10 10	8 10	10 13 10 10	9 10 10 10	8 10 11 11	8 8 8 8
8 8	10 11 10 10	10 11 10 9	10 10 9 10	10 10 10 10	8 10	10 12 10 10	10 10 10 11	10 10 11 11	8 6 6 6
7 8	10 11 10 10	10 11 10 9	10 10 10 10	10 10 10 10	8 10	10 12 10 10	11 10 10 10	10 10 11 10	6 7 7 7
7 8	10 11 10 10	10 11 10 9	10 10 10 10	10 10 10 10	8 10	10 11 10 10	10 10 10 10	10 10 10 10	7 7 7 7
7 8	10 11 10 10	10 11 10 6	10 10 10 10	10 10 10 10	8 9	9 11 10 10	10 10 10 9	10 10 10 10	7 7 7 7
8 8	10 10 10 10	10 11 10 6	10 10 10 10	10 10 10 10	8 9	9 10 10 10	9 9 8 8	10 10 10 10	7 7 10 10
7 8	10 10 10 10	10 11 10 6	10 10 10 10	10 10 10 10	7 9	9 10 10 10	8 8 8 8	10 10 9 9	10
7 8	10 10 10 9	10 11 10 6	10 10 10 9	10 9 10 10	7 9	9 10 10 10	8 8 6 8	9 9 9 9	
7 8	10 10 10 9	10 11 10 6	10 10 10 9	10 10 10 10	8 9	9 11 10 10	8 6 6 6	9 9 9 10	
7 8	10 10 10 9	10 11 10 10	10 10 10 10	10 10 10 10	7 9	9 11 10 10	6 6	10 12 12 12	
7 8	10 10 10 9	10 11 10 10	10 10 10 10	10 10 9 10	7 9	9 11 10 10		12 10 10 10	
7 8	10 10 10 9	10 11 10 10	10 10 10 10	9 10 9 11	7 10	9 11 10 10		10 12 12 12	
8 8	10 10 10 9	10 11 10 10	10 10 10 10	9 8 9 11	7 11	9 11 10 10		12 12 12 12	
8 8	10 10 10 9	10 11 10 10	9 10 10 10	9 9 9 11	6 11	10 10 10 10		12 12 11 11	
8 8	10 10 9 9	10 11 10 10	9 10 9 10	9 10 9 11	7 11	10 10 10 9		12 12 12 12	
8 8	10 10 9 9	10 11 10 10	9 9 9 10	9 10 9 11	7 11	10 10 9 10		12 12 12 12	
8 8	10 10 9 9	10 11 10 10	9 9 9 10	9 10 9 11	7 11	10 9 9 10		12 10 10 10	
7 8	10 10 9 9	10 11 10 10	9 9 9 10	9 10 9 11	6 11	10 9 9 10		10 10 10 10	
7 8	10 10 9 9	10 11 10 10	9 9 9 10	9 10 9 10	6 11	11 9 9 10		10 10 10 10	
7 8	10 10 9 9	10 11 10 10	9 9 9 10	9 10 9 10	7 11	11 9 10		10 10 10 10	
7 8	10 10 10 10	10 11 10 10	9 9 6 10	9 10 9 10	7 11	12 9 10		10 12 12 12	
7 8	10 10 10 10	10 11 10 10	10 9 6 10	9 10 9 10	10 10	13 9 10		12 12 13 13	
7 8	10 10 10 10	10 11 10 10	10 9 6 10	9 11 9 10	10 10	13 9 10		13 13 12 12	
	10 10 10 10	10 11 10 10	10 9 6 10	10 11 9 10	10 10	13 9 10		12 11 11 11	
	10 10 10 10	10 10 10 10	10 10 10 10	10 10 10 10				11 11 11 11	

Follicle Number

1	2	3	4	5	6	7	8	9	10
	101010	10101010 101010 9 9 9 9 9 9 9 1010 10101010 10 10	9 9 9 9 9 8 8 8 9 9 9 9 9 9 9 9 9	10101010 10101010 10 9 9 9 9 9 9 9 9 101010 10101010				11 11 11 11 11 101010 1010	
Sum									
422	1278	1381	1256	1376	559	1076	484	1368	370
No. of Cells									
55	129	138	133	141	63	106	54	130	47
Mean Cell heights									
7.7	9.9	10.0	9.4	9.6	8.9	10.2	9.0	10.5	7.9
Sum of squares									
3250	12686	13943	11960	13478	5105	11036	4456	14586	3104

Mean of mean cell heights of follicles = 9.5 units.

APPENDIX VIII

Thyroid Acinar Cell Heights of a Dairy Cow "Quiver"

Fed Thyroprotein (30 grams daily over 14 days)

Follicle Number

1	2	3	4	5	6	7	8	9	10
5 6 6 5	4 4 3	5 5 4 6	5 5 5 5	5 6 4 5	5 5 5 7	5 6	5 5 5	5 5 4 4	6 5 5 6
5 6 6 4	4 4 3	5 5 4 6	5 5 5 5	5 6 4 5	5 5 5 7	5 6	5 5 5	5 5 4 4	6 5 5 6
5 7 6 4	4 4 3	5 5 4 6	5 5 5 5	5 6 4 5	5 5 5 7	5 6	5 5 5	5 5 4 4	6 5 5 6
5 7 6 3	4 4 3	5 5 4 5	5 5 5 5	5 6 4 5	5 5 5 7	5 6	5 5 5	5 5 4 4	6 5 5 7
5 7 4 3	4 4 3	5 5 4 4	5 5 5 5	5 5 3 5	5 5 5 7	5 6	5 5 5	5 5 4 4	6 5 5 7
5 7 4 3	4 4 3	5 5 4 4	5 5 5 5	6 5 3 5	5 5 5 6	5 6	5 5 5	5 4 4 4	6 5 5 7
5 7 4 3	4 4 3	5 5 4 4	5 5 5 6	6 5 3 5	5 5 5 6	5 6	5 5 6	5 4 4 4	6 5 5 7
5 7 4 3	4 4 3	5 5 5 4	5 5 5 6	6 5 3 5	5 5 5 6	5 5	5 5 6	5 4 5 4	6 5 5 7
5 7 3 3	4 4 3	5 4 5 4	5 5 5 6	6 5 3 5	5 5 5 6	5 5	5 5 6	5 4 5 5	6 6 5 7
5 7 3 3	4 4 4	5 4 5 4	5 5 5 6	6 5 3 5	5 5 5 4	5 5	5 5 6	5 4 5 5	6 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	6 4 3 4	5 5 5 4	5 5	5 5 6	5 4 5 5	6 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	6 4 3 4	5 5 5 4	5 5	5 5 6	5 4 5 5	5 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	6 4 4 4	5 5 5 4	5 5	5 5 6	5 4 5 5	5 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	6 4 4 4	5 5 5 5	5 5	5 5 6	5 4 5 5	5 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	6 4 4 4	5 6 6 5	5 5	5 5 6	5 4 5 5	5 6 5 7
5 7 3 4	4 4 4	5 4 5 4	5 5 5 6	7 4 4 4	6 6 6 5	5 5	5 5 6	5 4 5 5	5 6 5 6
5 7 3 4	4 4 4	5 4 6 4	5 5 5 6	7 4 4 4	6 6 6 5	5 5	5 5 6	5 4 5 6	5 6 5 6
5 8 3 4	4 4 4	5 4 6 4	5 5 5 6	7 4 4 4	6 6 6 5	5 5	5 5 6	5 4 5 6	5 6 5 6
5 8 3 4	4 4 4	5 4 6 4	5 5 5 6	7 4 4 4	6 6 6 5	5 5	5 5	5 4 5 6	5 6 5 6
5 8 3 4	4 4 4	5 4 6 4	5 5 5 6	6 4 4 5	6 6 6 5	5 5	5 5	5 4 5 6	5 6 5 6
5 8 3 4	4 4 4	5 4 6	5 5 5 6	6 4 4 5	6 6 6 5	5 5	5 5	5 4 4 6	5 5 5 6
5 8 444	4 4 4	5 4 6	5 5 5 6	6 4 4 5	6 6 6	5 5	5 5	5 4 4 6	5 5 6 6
5 8 4	4 4 4	5 4 6	5 5 5 6	6 4 4 5	6 6 6	5 5	5 5	5 4 4 6	5 5 6 6
5 7 4	4 4 4	5 4 6	5 5 5 6	6 4 4 5	6 6 6	5	5 5	5 4 4 6	5 5 6 6
6 7 4	4 4 4	5 4 6	5 5 5 6	6 4 4 5	6 6 7		5 5	5 4 4 6	5 5 6 6
6 7 4	4 3 4	5 4 6	5 5 5 6	6 4 4 5	5 6 7		5 5	5 4 4 6	5 5 6 6
6 7 4	4 3	5 4 6	5 5 5 5	6 4 5 5	5 5 7		5 5	5 4 4	5 5 6 6
6 7 4	4 3	5 4 6	5 5 5 5	6 4 5	5 5 7		5 5	5 4 4	5 5 6 6

	Follicle Number									
	1	2	3	4	5	6	7	8	9	10
				5 5 5 5						6 5 5 5
				5 5 5 5						5 5 5 5
				5 5 5 5						5 5 5 5
				5 5 5 5						5 5 5 5
				5 5 5 5						5 5 5 5
				5 5 5 5						5 5 5 5
				5 5 5 5						
				5 5 5 5						
				5 5 5 5						
				5 5 5 5						
				5 5 5 5						
				5 5						
Sum	532	316	492	810	520	560	242	382	464	751
No. of Cells	106	82	104	158	110	102	47	74	100	148
Mean Cell Height	5.0	3.9	4.7	5.1	4.7	5.5	5.1	5.2	4.6	5.1
Sum of Squares	2920	1228	2870	4980	2566	3126	1252	1622	2196	3701

Mean of mean cell heights of follicles = 4·9 units

APPENDIX IX

Thyroid Acinar Cell Heights of a Dairy Cow "Unmask", Fed Thiourea
(25 grams daily over 21 days)

Follicle Number

1	2	3	4	5	6	7	8	9	10
35 35 35 35	24 24 25 25	20 20 20 20	30 30 30 30	25 25 25 25	24 24 24 24	24 24 24 24	22 22 22 22	20 20 20 20	20 20 20 20
32 35 35 35	25 25 26 26	20 20 23 23	30 30 30 30	25 25 25 25	23 23 23 20	25 25 25 25	24 24 24 24	20 20 22 22	20 20 20 20
32 35 24 25	26 28 26 26	23 23 23 20	20 30 30 30	24 24 24 24	20 20 22 22	25 25 24 24	24 24 26 26	23 23 20 20	20 20 20 20
33 26 25 25	24 24 24 24	20 20 20 20	30 30 30 30	24 24 24 24	22 24 24 24	24 20 20 20	26 26 26 23	20 20 20 20	20 20 20 20
34 20 20 20	22 22 22 22	22 22 22 18	30 30 30 30	24 23 23 23	24 24 22 20	20 20 20 20	25 25 25 25	18 18 18 18	20 20 20 21
34 22 22 22	24 24 23 23	18 18 18 24	30 30 30 30	23 21 21 21	20 20 20 22	20 20 20 20	25 25 25 23	18 18 18 18	21 22 22 22
34 22 22 23	23 20 20 20	24 24 25 30	30 30 30 30	21 19 19 19	22 22 22 23	20 18 18 18	23 23 23 23	18 10 10 10	22 22 24 24
34 23 23 23	20 7 7 8	30 32 32 32	30 30 30 28	20 20 20 20	25 25 25 27	20 20 20 20	22 22 22 22	10 10 12 12	24 24 24 25
36 25 25 24	8 8 8 10	34 34 34 34	28 28 28 28	20 20 19 19	27 27 27 28	22 22 22 22	22 22 22 26	12 12 15 15	25 25 22 20
36 24 20 18	10 10 10 10	34 36 36 36	26 26 26 24	20 24 24 24	30 30 30 30	24 24 24 24	26 26 26 28	15 15 15 15	20 20 20 20
38 18 18 22	30 30 30 30	36 34 34 32	24 24 24 24	24 20 20 20	28 28 30 30	28 28 28 28	28 28 28 26	15 20 20 20	20 20 18 18
38 22 22 22	25 25 25 25	32 34 34 34	24 24 23 23	18 18 18 18	32 32 32 32	28 26 26 26	26 26 24 24	20 20 18 18	18 18 18 18
38 22 24 25	25 25 24 24	30 30 32 32	23 23 23 23	22 22 23 23	35 35 35 33	26 26 25 25	24 24 24 24	18 18 18 18	18 19 19 19
36 25 25 25	24 24 24 23	32 32 30 33	20 20 20 20	23 23 22 22	33 32 32 32	25	24 24 24 24	18 20 20 20	19 19 19 19
36 25 25 24	23 23 23 20	33 33 24 24	20 20 20 20	20 20 20 20	32 32 28 28		26 26 26 25	20 20 20 18	20 20 20 20
35 24 24 25	20 20 20 20	20 20 20 20	20 18 18 18	20	28 28 28 28		25 25 25 25	18 18 18 18	22 22 22 22
32 28 28 25	20 20 20 24	20 24 24 24	18 18 18 18		25 25 25 25		25 25 25 25	16 16 16 16	22 25 25 25
32 25 25 25	24 24 24	24 20 20 20	18 20 25 25		27 27 27 27		25 26 26 26	17 17 17 17	25 25 24 24
32 25 25 30		18 18 18 19	25 28 28 28		30 32 32 32		26 26 28 28	18 18 18 18	23 23 23 23
32 30 30 30		19 19 19 19	28 28 28 28		32 31 31 31		28 28 28 30	20 20 20 20	22
32 30 30 32		18 18 18 18	28 28		31 30 30 30		30 31 31 31	21 21 21 21	
30 32 32 32		18 18 18 25			30 30 30 30		26 26 26 26	21 22 22 22	
31 32 32 28		27 27 27 27			28 28 28 26		25 25 25 25	22 24 24 24	

Follicle Number

1	2	3	4	5	6	7	8	9	10
28 27 27 27		27 27 27 29			26 26 24 24		25	24 24 24 22	
22 22 22 22		29 35 35 35			24			22 22 22 22	
20 20 20 24		35 35 32 32						22 21 21 21	
24 24 25 25		32 32						21 21	
25 23 20 20									
20 20 28 28									
28 35 35 36									
36 38 38 36									
36 34 34									
Sum									
3659	1521	2500	1177	1389	2632	1221	2367	2011	1623
Number of Cells									
131	71	97	50	64	97	53	94	107	77
Mean Cell Height									
27.9	21.4	25.8	23.5	21.7	27.1	23.0	25.2	18.8	21.0
Sum of Squares									
06209	35099	68914	28353	30493	72952	25319	60003	38949	34549

Mean of mean cell heights of follicles = 23·9 units.

APPENDIX X.

OBSERVATIONS ON ACINAR CELL HEIGHT.

(in Units; 1 unit = 2.25 μ)

NAME OF COW : QUARTZ. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	5	5	5	5	6	7	5	7	5	5
2	6	5	5	4	7	6	6	6	4	6
3	5	4	6	4	7	5	7	8	5	6
4	7	5	5	5	6	6	6	6	6	5
5	6	6	5	5	7	4	6	5	5	6
6	5	5	6	5	7	7	7	7	5	5
7	5	4	5	4	7	6	5	6	6	5
8	5	4	5	5	6	5	6	5	6	6
TOTAL	44	38	42	37	53	46	48	50	42	44
	5.5	4.8	5.3	4.6	6.6	5.8	6.0	6.3	5.3	5.5

Mean Acinar cell height of the gland = 5.57.

APPENDIX NO. XI.

OBSERVATIONS ON ACINAR CELL HEIGHT,

(in Units ; 1 unit = 2.25 μ).

NAME OF COW : LARKSPUR. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	10	10	10	9	8	10	8	8	8	7
2	8	11	10	9	7	8	8	9	10	10
3	8	9	11	10	8	10	8	8	12	10
4	7	11	9	8	9	9	10	7	10	10
5	9	10	8	10	10	9	7	8	11	7
6	8	13	11	9	10	8	9	8	9	9
7	8	11	10	9	9	8	8	9	8	9
8	9	10	11	8	10	8	9	8	10	9
TOTAL	67	85	80	72	71	70	67	65	78	71
	8.4	10.6	10.0	9.0	9.0	8.8	8.4	8.1	9.6	9.0

Mean Acinar cell height of the gland = 9.09.

APPENDIX XII.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units; 1 Unit = 2.5 μ).
TREATMENT : NORMAL.

NAME : No. 67.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1	6	7	6	6	7	6	5	7	5	6
2	6	7	5	6	6	5	7	7	6	7
3	5	5	5	6	7	5	6	6	7	7
4	5	6	6	6	5	5	6	7	6	8
5	5	6	6	6	4	6	5	7	7	6
6	7	7	5	6	6	5	5	6	6	7
7	5	7	6	5	6	5	6	7	6	6
8	7	6	6	5	5	5	5	7	6	6
TOTAL	46	51	45	46	46	42	45	52	49	53
	5.8	6.4	5.6	5.8	5.8	5.3	5.6	6.5	6.1	6.6

Mean acinar cell height of the gland = 5.95.

APPENDIX NO. XIII.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units; 1 unit = 2.5 μ)
NAME OF COW : No. 84. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1	7	8	10	7	9	7	6	9	10	8
2	7	8	10	6	9	7	5	8	9	10
3	7	7	7	7	8	6	7	8	9	8
4	8	9	7	8	7	7	8	7	8	7
5	8	8	11	8	7	5	7	7	9	7
6	8	7	8	8	8	6	6	8	7	8
7	8	9	7	8	6	6	6	7	6	10
8	7	8	7	7	5	7	6	8	10	10
TOTAL	60	64	67	59	59	51	51	62	68	68
	7.5	8.0	8.4	7.4	7.4	6.4	6.4	7.6	8.5	8.5

Mean acinar cell height of the glad = 7.59

APPENDIX NO. XIV.

OBSERVATIONS ON ACINAR CELL HEIGHT (In Units; 1 unit = 2.5 μ).

NAME : No. 24. 59 TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	5	5	5	7	6	6	5	8	7	10.
2	6	6	6	6	5	6	6	7	6	8
3	6	5	5	5	7	5	8	6	6	7
4	5	5	6	7	6	7	7	6	8	7
5	6	5	7	6	6	6	7	7	8	7
6	5	5	5	6	7	6	8	7	7	8
7	6	6	6	7	8	7	7	6	8	7
8	5	5	5	6	9	6	6	6	7	7
TOTAL	44	42	45	50	54	49	54	53	57	61
	5.5	5.3	5.6	6.3	6.8	6.1	6.8	6.6	7.1	7.6

Mean acinar cell height of the gland = 6.37

APPENDIX NO. XV.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units; 1 Unit = 2.5 μ)

NAME : NO. 16. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	6	6	10	7	8	5	10	7	6	7
2	6	6	6	7	7	7	9	7	6	8
3	6	7	7	6	9	6	10	6	8	9
4	7	7	7	6	6	7	7	7	9	9
5	7	7	7	7	7	7	8	7	8	8
6	8	6	6	7	6	6	7	7	8	8
7	8	6	6	6	7	6	8	8	8	9
8	6	7	7	6	7	7	7	8	8	8
TOTAL	54	52	56	52	57	51	66	57	61	66
	6.8	6.5	7.0	6.5	7.1	6.4	8.2	7.1	7.6	8.2

Mean acinar cell height of the gland = 7.14.

APPENDIX NO. XVI.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units ; 1 unit = 1.25 µ).

NAME : PAGAN. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1	10	6	7	7	7	7	8	7	8	10
2	10	7	7	6	7	8	10	10	7	10
3	7	7	7	7	8	7	8	8	6	9
4	9	6	7	7	9	7	9	9	6	9
5	8	7	7	7	8	9	8	7	7	8
6	6	7	6	7	7	10	9	8	7	7
7	10	7	7	7	9	7	7	6	6	9
8	10	6	7	7	7	8	8	9	7	8
TOTAL	70	63	55	55	62	63	67	64	54	70
	8.8	7.9	6.9	6.9	7.8	7.8	8.4	8.0	6.8	8.8

Mean acinar cell height of the gland = 7.81

APPENDIX NO. XVII.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units ; 1 unit = 1.25).

NAME : TRAMP. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1	6	7	5	7	7	8	6	7	6	6
2	6	7	5	7	7	8	6	7	6	7
3	7	6	6	6	6	7	6	7	6	7
4	7	6	6	6	6	8	7	6	7	7
5	6	7	5	7	6	6	7	6	7	7
6	6	7	6	7	7	7	6	6	7	7
7	6	7	6	6	6	7	6	7	6	7
8	7	7	5	6	7	8	7	7	6	7
TOTAL	51	54	44	52	52	59	51	53	51	55
	6.4	6.8	5.5	6.5	6.5	7.4	6.4	6.6	6.4	6.9

Mean acinar hell height of the gland = 6.54

APPENDIX NO. XVIII.

OBSERVATIONS ON ACINAR CELL HEIGHT (in units; 1 unit = 2.5 μ).

NAME : TIKI. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1.	6	7	6	5	7	6	7	7	5	7
2.	6	7	5	5	7	5	6	6	6	7
3.	6	6	5	6	6	7	7	5	6	8
4.	6	7	6	6	5	8	6	6	7	6
5.	6	7	6	6	6	6	7	5	7	7
6.	6	6	7	7	5	7	6	6	6	7
7.	7	7	5	6	7	6	7	6	5	6
8.	6	7	6	6	7	7	7	5	6	6
TOTAL	49	60	46	47	50	52	53	46	48	54
	6.1	7.5	5.8	5.9	6.3	6.5	6.6	5.8	6.0	6.8

Mean acinar cell height of the gland = 6.33.

APPENDIX NO. XIX.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units; 1 unit = 2.5 μ).

NAME : PHLOX. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1.	6	9	7	10	6	6	8	7	6	10
2.	6	10	8	8	7	8	9	7	10	9
3.	7	7	7	9	5	9	10	6	10	9
4.	6	10	9	8	8	7	9	7	9	10
5.	6	8	7	10	4	9	7	7	10	7
6.	7	7	8	8	6	7	6	9	7	9
7.	8	9	8	8	7	5	7	8	6	9
8.	7	10	8	9	8	8	10	9	9	8
TOTAL	53	70	62	70	51	59	66	60	67	71
	6.6	8.8	7.8	8.8	6.4	7.4	8.3	7.5	8.4	8.9

Mean acinar cell height of the gland = 7.89.

APPENDIX NO. XX.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units: 1 unit = 1.5 μ).

NAME : SQUIRE. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1.	6	5	6	6	7	7	6	7	7	7
2.	6	6	5	5	6	7	7	6	5	6
3.	5	6	4	7	7	6	6	6	6	8
4.	6	5	5	6	6	5	7	5	5	7
5.	5	5	4	7	7	4	6	7	7	6
6.	6	6	5	6	5	5	7	7	7	5
7.	5	5	5	7	6	5	6	7	6	8
8.	5	5	4	6	5	6	5	6	5	7
TOTAL	:44	43	38	50	49	45	50	51	48	54
	5.5	5.4	4.8	6.3	6.1	5.6	6.2	6.4	6.0	6.8

Mean acinar cell height of the gland = 5.91

APPENDIX NO. XXI.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units: 1 unit = 1.5 μ).

NAME : No. 51. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1.	10	7	10	8	10	7	10	8	9	9
2.	8	8	10	7	9	6	10	9	9	8
3.	10	8	11	7	7	8	8	11	8	9
4.	10	9	9	8	6	8	8	11	6	9
5.	10	8	8	9	7	7	10	11	9	8
6.	8	8	10	8	9	7	8	7	9	7
7.	8	10	11	9	10	8	10	9	7	10
8.	8	9	9	8	6	7	9	8	8	8
TOTAL	:72	67	78	64	64	58	73	74	65	68
	9.0	8.4	9.8	8.0	8.0	7.3	9.1	9.3	8.1	8.5

Mean acinar cell height of the gland = 8.55.

APPENDIX NO. XXII.

OBSERVATIONS ON ACINAR CELL HEIGHT (in Units; 1 unit = 2.5μ).

NAME : No. 19. TREATMENT : NORMAL.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	6	10	7	7	6	9	6	6	8	7
2.	6	10	7	8	5	8	5	7	7	7
3.	6	7	7	6	6	9	6	7	7	6
4.	10	7	8	6	7	4	7	6	5	6
5.	9	7	6	5	5	8	7	6	8	7
6.	7	7	6	7	7	4	7	6	6	7
7.	9	7	8	5	7	6	8	5	7	6
8.	7	6	7	7	6	5	6	6	5	5
TOTAL	60	61	57	51	49	53	52	49	53	51
	7.5	7.6	7.1	6.4	6.1	6.6	6.5	6.1	6.6	6.4

Mean acinar cell height of the gland = 6.69.

APPENDIX XXIII.

Observations on Acinar Cell Height
(in Units; 1 unit = 2.5 μ).

NAME OF COW : MONA. TREATMENT : Thyroprotein 10 grams daily over 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	5	3	3	5	4	3	5	5	4	4
2	5	4	3	5	3	3	5	5	4	5
3	5	4	4	4	3	4	4	5	2	5
4	6	5	3	5	4	3	4	4	3	5
5	5	4	3	3	5	3	5	4	3	5
6	6	4	4	5	3	4	4	4	4	4
7	6	5	3	4	4	3	4	4	2	4
8	7	4	4	4	4	3	4	5	3	4
TOTAL	45	33	27	35	30	26	35	36	25	36
	5.6	4.1	3.4	4.4	3.8	3.3	4.4	4.5	3.1	4.5

Mean Acinar cell height of the gland = 4.11.

APPENDIX XXIV.

Observations on Acinar Cell Height
(in Units; 1 unit = 2.5 μ).

NAME OF COW : NELL. TREATMENT : Thyroprotein 10 grams daily over 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	5	5	6	5	5	7	6	5	5	5
2	6	4	6	5	6	6	6	5	5	5
3	5	4	5	7	6	7	5	4	4	4
4	5	5	6	6	5	8	6	5	4	5
5	4	5	5	6	6	6	5	4	5	5
6	5	3	5	6	5	8	5	6	5	4
7	5	4	4	6	5	8	6	5	4	3
8	4	4	5	5	6	6	6	4	5	5
TOTAL	39	34	42	46	44	56	45	38	37	36
	4.9	4.3	5.3	5.8	5.5	7.0	5.6	4.8	4.6	4.5

Mean Acinar cell height of the gland = 5.23.

APPENDIX NO. XXV.

Observations on Acinar Cell Height
(in units; 1 unit = 2.5 µ).

NAME OF COW : MANDOLIN. TREATMENT : 20 grams Thyroprotein daily over 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	4	5	4	3	4	4	2	5	6	4
2	4	4	3	3	3	4	3	3	5	5
3	5	2	5	2	3	4	2	3	4	4
4	3	3	4	3	2	3	2	3	5	4
5	2	2	3	4	2	3	3	3	3	4
6	4	3	3	3	3	2	3	3	4	6
7	3	3	3	3	3	3	5	3	3	5
8	3	2	4	3	3	3	3	3	4	4
TOTAL	28	24	29	24	23	26	23	26	34	36
	3.5	3.0	3.6	3.0	2.9	3.3	2.9	3.3	4.3	4.5

Mean acinar cell height of the gland = 3.43.

APPENDIX NO. XXVI.

Observations on Acinar Cell Height,
(in units; 1 unit = 2.5 µ).

NAME OF COW : LILAC. TREATMENT : 30 grams Thyroprotein daily over 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	4	5	4	3	4	3	4	4	4	4
2	3	4	3	4	4	3	5	5	4	4
3	4	4	5	3	5	2	3	3	4	5
4	4	4	5	3	4	3	5	2	3	4
5	3	5	6	3	5	6	4	3	5	4
6	4	4	5	4	6	4	3	2	4	3
7	3	4	4	3	4	4	4	2	4	2
8	4	4	5	3	7	3	3	3	3	2
TOTAL	29	34	37	26	39	28	31	24	31	28
	3.6	4.3	4.6	3.3	4.9	3.5	3.8	3.0	3.8	3.5

Mean acinar cell height of the gland = 3.83.

APPENDIX NO. XXVII.

Observations on Acinar Cell Height,
(in units; 1 unit = 2.5 μ).

NAME OF COW : SOPHIE. TREATMENT : 30 grams Thyroprotein daily over 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	4	5	5	5	5	5	5	3	2	4
2	4	4	3	5	4	4	5	3	3	4
3	3	4	2	4	3	5	4	2	3	3
4	4	3	2	4	3	5	4	3	2	3
5	5	4	2	3	2	4	5	3	4	3
6	3	3	3	2	3	2	3	3	3	2
7	4	3	3	3	2	4	5	3	4	3
8	4	3	2	2	2	3	4	3	4	2
TOTAL	31	29	22	28	24	32	35	23	25	24
	3.9	3.6	2.8	3.5	3.0	4.0	4.4	2.9	3.1	3.0

Mean acinar cell height of the gland = 3.42

APPENDIX NO. XXVIII

Observations on Acinar Cell Height.
(in units; 1 unit = 2.5 μ).

NAME OF COW : ULEX. TREATMENT : 10 grams Thyroprotein daily over 56 days.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10
1	4	5	4	5	3	3	5	5	4	4
2	4	4	5	5	3	3	5	4	5	5
3	3	5	3	5	3	4	4	5	5	5
4	4	5	3	3	2	3	4	5	4	4
5	4	4	4	4	3	2	4	4	4	3
6	4	4	3	4	3	3	3	4	5	3
7	3	5	4	4	2	2	5	4	4	3
8	4	5	4	3	3	2	4	4	4	3
TOTAL	30	37	30	33	22	22	34	35	35	30
	3.8	4.6	3.8	4.1	2.8	2.8	4.3	4.4	4.4	3.8

Mean acinar cell height of the gland = 3.88

APPENDIX NO. XXIX.

(Observations on Acinar Cell Height, in units; 1 unit = 1.5 μ).

NAME OF COW : NORA. TREATMENT : 10 grams Thyroprotein daily over 56 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	3	2	3	3	3	3	3	3	5	5
2	3	3	2	3	2	3	4	3	4	4
3	3	3	2	2	2	4	5	4	5	5
4	3	2	3	3	3	4	4	3	4	5
5	3	3	3	2	3	3	4	5	4	3
6	3	4	3	2	4	4	5	4	5	4
7	3	4	3	3	3	4	3	5	3	4
8	3	2	3	2	4	3	4	3	4	3
TOTAL	24	23	22	20	24	28	32	30	34	33
	3.0	2.9	2.8	2.5	3.0	3.5	4.0	3.8	4.3	4.1

Mean acinar cell height of the gland = 3.39.

APPENDIX NO. XXX.

(Observations on Acinar cell Height, in units; 1 unit = 1.5 μ).

NAME OF COW : TINDER. TREATMENT : Thyroprotein fed up to 76 days before killing.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	14	11	7	10	10	10	15	10	9	13
2	13	11	10	14	10	10	11	8	8	13
3	13	11	9	9	13	9	14	10	9	13
4	11	12	10	9	9	10	12	8	9	12
5	13	15	7	10	10	9	9	9	11	15
6	13	15	9	12	9	11	13	9	12	11
7	12	11	9	13	11	12	13	11	12	12
8	13	10	10	9	9	11	10	9	11	12
TOTAL	102	96	71	86	81	82	97	82	81	101
Y	12.7	12.0	8.9	10.7	10.1	10.3	12.1	10.3	10.1	12.6

Mean acinar cell height of the gland = 10.98.

APPENDIX NO. XXXI.

(Observations on Acinar Cell Height, in units; 1 unit = 2.5 μ).

NAME OF COW : UPSTART. TREATMENT : Thyroprotein fed up to 76 days before killing.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	9	9	8	9	8	10	7	6	6	7
2	8	9	9	8	7	9	6	7	6	7
3	9	9	9	9	7	9	8	7	7	8
4	9	9	8	9	8	8	8	6	8	7
5	7	8	9	8	7	10	8	7	7	9
6	9	10	8	9	10	7	9	7	7	8
7	9	10	9	7	9	8	7	7	6	9
8	9	9	9	7	8	7	7	6	6	9
TOTAL	69	73	69	66	64	68	60	53	53	64
	8.6	9.1	8.6	8.3	8.0	8.5	7.5	6.6	6.6	8.0

Mean acinar cell height of the gland = 8.00

APPENDIX NO. XXXII.

(Observations on Acinar cell height, in units; 1 unit = 2.5 μ).

NAME OF COW : QUIRE TREATMENT : Thyroprotein fed up to 76 days before killing.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	5	6	5	4	5	5	6	5	4	5
2	6	7	4	4	5	4	7	4	3	5
3	7	7	4	3	5	4	7	3	4	4
4	6	5	4	4	4	5	7	4	7	5
5	5	6	5	4	5	5	5	4	7	4
6	5	5	5	5	5	5	5	5	4	4
7	5	6	5	6	5	4	4	5	3	5
8	6	6	4	5	5	6	6	5	5	4
TOTAL	45	48	36	35	39	38	47	35	37	36
	5.6	6.0	4.5	4.4	4.9	4.8	5.9	4.4	4.6	4.5

Mean acinar cell height of the gland = 4.96

APPENDIX NO. XXXIV.

(Observations on acinar cell height, in units; 1 unit = 2.5 μ).

NAME OF COW : RAUPO. TREATMENT : Thyroprotein fed up to 76 days before killing.

CELL NO.	FOLLICLE NUMBER.									
	1	2	3	4	5	6	7	8	9	10
1	8	10	10	11	10	10	10	10	10	8
2	8	8	11	10	7	9	9	11	8	7
3	9	10	9	9	8	7	7	7	7	6
4	7	8	10	8	8	10	7	11	8	8
5	10	9	8	9	10	10	10	8	7	8
6	10	10	9	8	8	8	8	9	7	7
7	8	11	8	7	7	7	7	10	8	9
8	7	9	8	10	8	9	8	8	9	8
TOTAL	67	75	73	72	66	70	66	64	64	61
	8.4	9.4	9.1	9.0	8.3	8.8	8.3	8.0	8.0	7.6

Mean acinar cell height of the gland = 8.49.

APPENDIX NO. XXXV .

(Observations on acinar cell height, in units; 1 unit = 2.5 μ).

NAME OF COW : ROBBIE. TREATMENT : Thyroprotein fed up to 76 days before killing.

CELL NO.	FOLLICLE NUMBER									
	1	2	3	4	5	6	7	8	9	10
1	12	10	10	7	7	7	9	8	7	7
2	13	7	10	8	9	9	8	6	7	8
3	8	9	12	7	8	9	7	8	10	9
4	7	9	10	10	8	8	8	10	10	8
5	8	8	9	8	9	9	8	8	10	12
6	9	7	9	7	8	7	7	9	7	9
7	11	10	9	9	10	9	8	7	9	10
8	9	9	9	10	9	10	7	8	6	8
TOTAL	77	69	78	66	68	68	62	64	66	71
	9.6	8.6	9.8	8.3	3.5	8.5	7.8	8.0	8.3	8.9

Mean acinar cell height of the gland = 8.63.

APPENDIX NO. XXXVI.
(Observations on acinar cell height,
in units; 1 unit = 2.5 μ).

NAME : QUACK.

TREATMENT : 21 days (Thiourea 10 grams 7 days, 15 grams 14 days.

CELL NO.	FOLLICLE NUMBER 1	2	3	4	5	6	7	8	9	10
1	15	15	15	18	15	15	12	18	15	15
2	15	15	14	19	15	15	12	17	15	16
3	17	16	10	17	14	16	13	18	18	20
4	15	16	9	18	15	17	13	20	18	17
5	17	15	10	16	14	15	12	20	20	22
6	16	16	10	10	13	14	12	17	22	17
7	14	17	12	12	10	13	12	18	20	15
8	15	17	12	12	11	13	13	16	18	17
9	15	18	15	12	12	14	14	17	15	20
10	16	16	14	13	15	14	11	18	17	20
11	15	19	12	15	12	13	11	18	15	21
12	15	17	14	15	13	10	12	15	18	19
13	15	20	13	13	15	11	11	17	22	17
14	14	17	15	15	15	15	15	16	16	17
15	15	16	13	25	13	12	16	16	20	18
16	16	17	15	20	10	16	15	17	19	21
17	14	15	14	15	12	15	15	17	20	16
18	13	15	12	16	16	15	15	18	15	19
19	14	15	13	15	12	15	16	17	17	18
20	15	16	10	15	13	10	17	15	17	22
TOTAL	301	328	252	311	265	278	267	345	357	367
	15.1	16.4	12.6	15.6	13.3	13.9	13.4	17.3	17.9	18.4

Mean acinar cell height of the gland = 15.39.

APPENDIX NO. XXXVII

(Observations on acinar cell height,
in units; 1 unit = 2.5μ).

NAME : UNION. TREATMENT : (Thiourea 10 grams, 7 days, 15 grams, 14 days.

CELL NO.	FOLLICLE NUMBER. 1	2	3	4	5	6	7	8	9	10.
1	20	15	15	17	17	17	25	14	22	20
2	20	18	15	19	17	17	24	14	21	20
3	20	18	16	20	20	19	20	17	20	22
4	22	15	18	21	22	17	20	17	20	18
5	22	18	18	21	22	18	24	20	20	18
6	22	9	18	18	20	20	23	15	20	18
7	23	15	20	21	22	21	19	15	24	19
8	22	18	20	19	17	21	24	14	24	16
9	22	17	18	22	18	21	19	13	20	20
10	22	20	20	20	23	23	20	17	17	21
11	23	20	18	20	21	17	24	17	18	17
12	22	17	19	20	20	18	24	13	20	15
13	24	18	19	19	17	18	20	11	22	19
14	26	20	19	20	16	18	19	17	20	18
15	22	20	20	20	15	17	20	11	16	23
16	23	18	20	20	17	20	17	17	17	20
17	24	18	19	22	19	21	17	20	17	19
18	25	20	18	22	19	21	18	20	17	19
19	24	19	18	24	21	21	22	15	18	20
20	24	16	19	19	21	21	20	16	20	23
TOTAL	452	349	367	404	384	386	419	313	393	385
	22.6	17.5	18.4	20.2	19.2	19.3	20.9	15.6	19.6	19.2

Mean acinar cell height of the gland = 19.25.

APPENDIX NO. XXXVIII.

(Observations on acinar cell height, in units; 1 unit = 2.5 μ).

NAME : UPRIGHT. TREATMENT : Thiourea 25 grams over 21 days.

CELL	FOLLICLE NUMBER									
NO.	1	2	3	4	5	6	7	8	9	10
1.	20	25	20	19	20	20	24	21	25	22
2	21	26	27	20	21	22	21	25	26	23
3	22	26	26	25	27	24	26	24	22	21
4	22	26	23	25	24	25	26	27	27	28
5	21	25	24	26	27	28	26	27	29	30
6	20	26	31	24	23	22	21	23	26	24
7	20	25	26	27	28	29	30	23	24	21
8	19	27	24	26	25	25	29	23	25	27
9	20	27	27	28	24	22	25	24	21	25
10	20	26	27	24	23	22	24	25	24	23
11	24	24	26	22	23	22	26	20	26	29
12	24	25	26	27	21	23	27	26	24	23
13	21	24	21	20	23	22	24	24	25	24
14	23	26	25	23	23	22	25	27	24	26
15	23	27	23	26	28	25	28	26	27	25
16	20	26	23	27	26	21	24	20	19	24
17	21	22	22	24	25	27	22	21	24	25
18	23	25	26	24	25	20	22	24	25	26
19	23	24	27	22	23	26	25	24	22	21
20	24	25	26	27	28	27	24	25	30	23
TOTAL	431	507	500	486	487	474	499	479	496	490
	21.6	25.4	25.0	24.3	24.4	23.7	24.9	23.9	24.8	24.5

Mean acinar cell height of the gland = 24.25.