

Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

A STUDY OF CHANGES IN THE THICKNESS AND
CHEMICAL COMPOSITION OF THE SKIN OF
SHEEP DURING GROWTH AND AFTER SHEARING

by

MARIA MANIKA WODZICKA

Being a thesis presented in partial fulfillment
of the requirements for the
degree of M. Agr. Sc.

Massey Agricultural College,
University of New Zealand.

March, 1954.

A C K N O W L E D G E M E N T S

I wish to express my gratitude to members of the Ruakura Animal Research Station and the Massey Agricultural College staffs for the help I have received from them during this investigation.

Special thanks are due to my supervisors Professor A.L. Rae and Dr. C.P. McMeekan; to Mr. W.G. Whitlestone, Mr. N.T. Clare, Miss P. Bartram, Dr. J. James, Mr. F.R. Cockrem and Mr. P.M. Linklater for help and advice.

For technical assistance I am indebted to Mrs. R. Holmberg, Mr. D. Somerville, Mr. J. Kerr, Mr. B. Miller, Mr. J. Gibson and Mr. B. Wright.

This thesis was carried out under the tenure of the Shell Scholarship.

TABLE OF CONTENTS

	<u>Page</u>
<u>GENERAL INTRODUCTION</u>	1
<u>CHAPTER I - DEVELOPMENT OF TECHNIQUES</u>	4
<u>A. SKIN THICKNESS MEASUREMENT</u>	
INTRODUCTION	4
I. METHODS AND MATERIALS	5
(a) Instruments	5
(1) Instrument for measurement of skin thickness	5
(ii) Methods of making frozen sections	8
(iii) Other instruments and materials.	8
(b) Experimental Animals	9
II. NATURE AND SCOPE OF EXPERIMENTS	9
(a) Preliminary Experiment I	11
(1) Introduction	11
(2) Plan of Experiment	12
(3) Results	15
(b) Preliminary Experiment II	22
(1) Introduction	22
(2) Plan of Experiment.	23
(3) Results	23
(4) Sampling procedure finally adopted	26
DISCUSSION	27
SUMMARY	29

Page

B. <u>CHEMICAL ANALYSIS</u>	30
INTRODUCTION	30
I. METHODS AND MATERIALS	31
(a) Apparatus	31
(b) Sampling Procedure and Analysis	32
II. NATURE AND SCOPE OF EXPERIMENTS	34
SUMMARY	44

<u>CHAPTER II</u> - DISTRIBUTION OF SKIN THICKNESS OVER THE BODY OF SHEEP	45
INTRODUCTION	45
I. METHODS AND MATERIALS	45
(a) Outline of Experiment	45
(b) Experimental Animals	46
(c) Technique	46
II. RESULTS	47
DISCUSSION	47
SUMMARY	49

<u>CHAPTER III</u> - THE SKIN DEVELOPMENT EXPERIMENT	50
INTRODUCTION	50
I. METHODS AND MATERIALS	50
(a) Plan of Experiment	50
(b) Experimental Animals	52
(c) Experimental Technique	58
(d) Sampling Procedure	59

	<u>Page</u>
II. RESULTS	60
(a) Skin Thickness	60
(b) Chemical Composition	71
DISCUSSION	81
SUMMARY	85
<u>CHAPTER IV - THE SHEARING EXPERIMENT</u>	86
INTRODUCTION	86
I. METHODS AND MATERIALS	86
(a) Plan of Experiment	86
(b) Experimental Animals	89
(c) Experimental Technique	89
II. RESULTS	90
(a) Skin Thickness	90
(b) Chemical Composition	93
DISCUSSION	94
SUMMARY	96
<u>GENERAL SUMMARY AND DISCUSSION</u>	97

GENERAL INTRODUCTION

Studies of the histology, physiology and biochemistry of the skin of sheep are of interest not only because of the economic value of leather and wool but also because skin is the first tissue to come in direct contact with the environment and hence plays an important part in such vital processes as heat regulation and defence against virus and bacterial invasion.

Experiments described in this thesis were designed to investigate the problem of the tearing of lambs' pelts which occurs in the New Zealand Freezing Works. It is found that some pelts of both the New Zealand Romney Marsh * and the New Zealand Romney Marsh-Southdown cross fat lambs are so thin that during the flaying and fellmongering processes they tear or else become stretched (the latter defect being called the "butcher's strain" fault). The incidence of these two faults is not correlated with the peak employment of unskilled labour in the Freezing Works, but occurs sporadically throughout the season. Therefore, whilst careless butchering and the faulty use of chemicals in the fellmongeries could be responsible for part of the trouble encountered, it seemed likely that an inherent skinweakness might also exist.

* The term "New Zealand Romney" is used here to indicate that the Romney sheep studied and henceforth referred to in this work as "Romney", were grade sheep, the offspring for many generations of Romney Marsh rams.

Many workers have studied the histological and chemical basis of pelt and leather defects in the pelt at the end of the fellmongering process and in the finished leather product. The limitations of this approach are, however, considerable. The identity and past history of the animal are as a rule unknown and hence it is frequently difficult to ascertain whether the defects in the product are due to faulty butchering, fellmongering or tanning or to weaknesses in the living skin. It is believed by the author that a fruitful line of approach lies in studying the conditions during the life of the animal which may cause skin weaknesses and therefore lead to leather defects. This was the approach adopted in this thesis.

An instrument was developed for the measurement of skin thickness because thinness is an outstanding characteristic of the torn pelts. Techniques were adapted for the analysis of the protein, fat and water content of the skin since these are the three main constituents of skin present in sufficient amounts to be measurable by ordinary chemical methods in small samples. The protein content of skin is of interest in addition, because leather is made from the protein fraction.

Changes in the thickness and chemical composition of the skin of 12 Romney wether lambs were followed from within ten days of birth to five months of age because it was considered desirable, before studying abnormalities, to study these changes as they normally occurred with increasing age.

Since the Freezing Works will not usually accept sheep for slaughter unless they have been shorn three weeks previously, and since most sheep are shorn once a year, a study was also made of the effect of shearing on the thickness and chemical composition of the skin of 16 Romney ewes.

Thus the initial problems in this approach were examined and the basis was laid down for future work on these lines.

C H A P T E R I

DEVELOPMENT OF TECHNIQUES

A. SKIN THICKNESS MEASUREMENT.

INTRODUCTION:

Before any investigation concerning changes in skin thickness during the growth of a lamb and in its adult life could be carried out, it was necessary to develop a technique for the rapid and accurate measurement of skin thickness. This involved not only the design of an instrument which would measure accurately skin thickness, but also a sampling method which would ensure:-

- (1) Removal of the skin from the animal in such a way that comparable cell layers of skin were taken off at each sampling. Moreover, it was necessary that these layers correspond with those removed with the pelt by flaying.
- (2) Removal of the wool without altering the skin thickness, cell structure and chemical composition.

It also seemed desirable to find a method for preserving the skin without altering either its thickness or its chemical composition. This would enable thickness measurements and chemical analyses to be made the day after the samples were taken and permit the sampling of larger numbers of animals per day.

I METHODS AND MATERIALS.

(a) Instruments.

(1) The instrument for measurement of skin thickness.

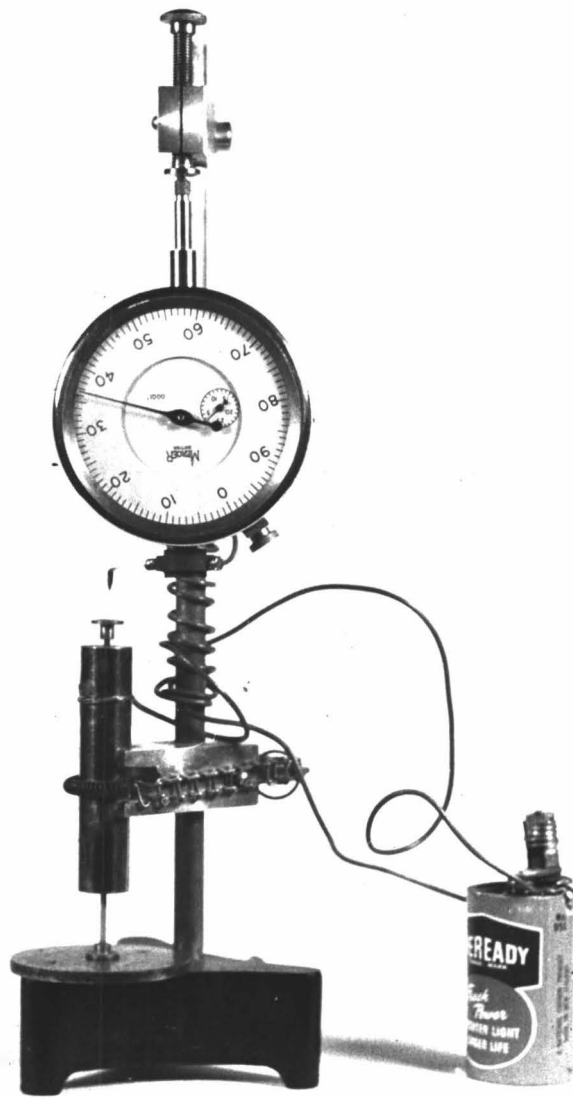
Nicov (1931) and Carstens and Kinzelbach (1933) attempted to measure skin thickness in cattle without taking the skin off the animal. Their method consisted of picking up a fold of skin, measuring its thickness under a constant pressure and dividing the result by two, to obtain the thickness of the skin. No repeatability figures are given, nor have the authors determined what this measurement represents histologically. Thus, it is unknown whether measurements by this method include some subcutaneous tissue as well as the epidermis and the dermis, and whether the amount measured varies between measurements and between animals.

Clarke, Stuart and Frey (1937) measured the thickness of "partly cured lamb skins with the microscope used on sections cut for histological examination." They give no further details of their curing and histological techniques. Since they did not measure fresh skin it is impossible to determine the extent of the changes in skin thickness resulting from the curing and the pre-section cutting treatment. They did not obtain any repeat measurements of the same sample in order to determine the accuracy of the method. Their method moreover involves considerable histological work and therefore, the number of samples which can be measured is limited.

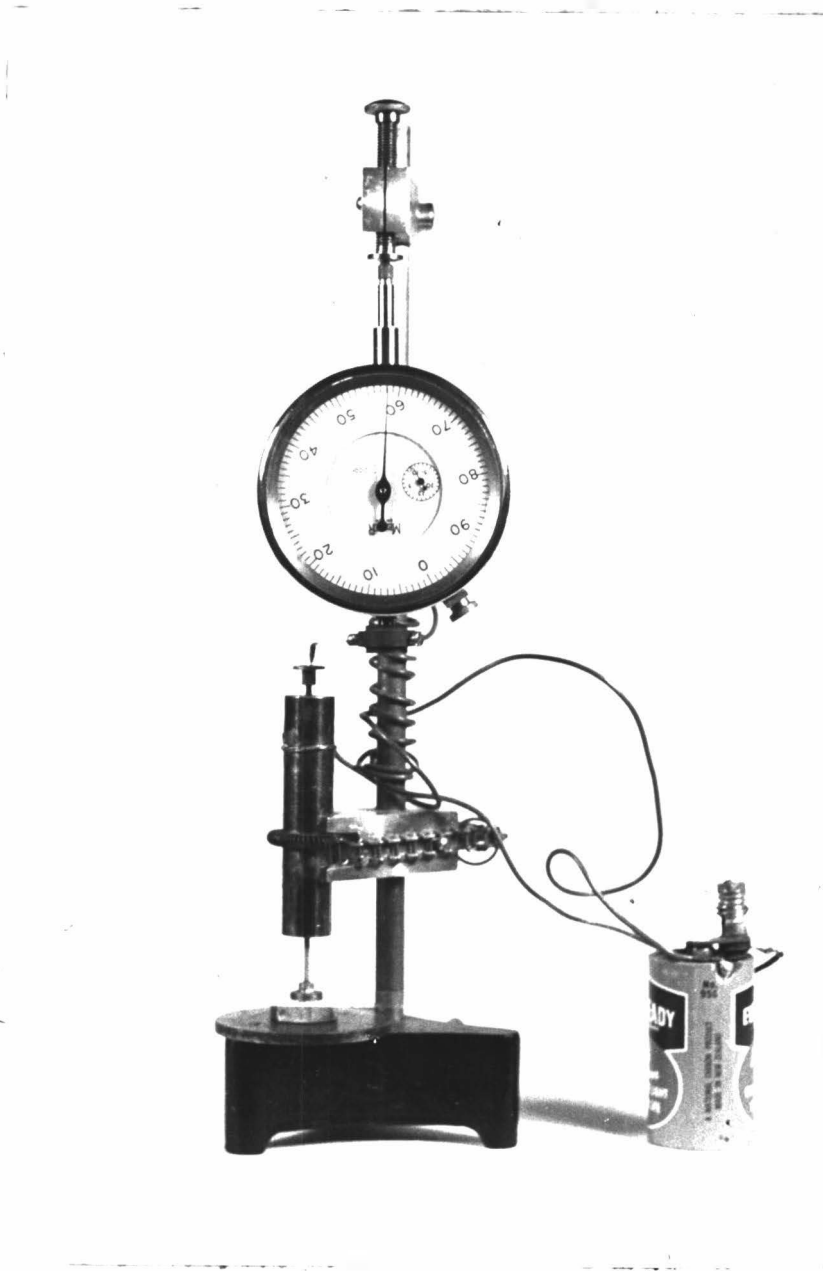
For the present investigation, an instrument was required which measured skin thickness quickly and accurately. The histological techniques involving section cutting were considered to be too time consuming whilst the "pinch" methods described above needed an objective measurement method for comparison before they could be tested and possibly accepted as valid means for the measurement of skin thickness. A new instrument was therefore designed in which skin thickness was measured under a small constant pressure. Photographs 1, 2 and 3 illustrate the instrument. The piece of skin to be measured (after being removed from the animal and cleared of wool by the methods described in sections I (a) (3) and II (b) (4) of this chapter) was placed under a constant weight disc G. The thickness value was recorded on a Mercer spring gauge I, read when the indicator lamp P flickered indicating a closed circuit. The difference between the gauge readings when the sample was under G and when it was absent, measured the thickness of the skin. This instrument for the measurement of skin thickness will henceforth be referred to as the "Instrument". Its detailed description follows below.

Detailed description:

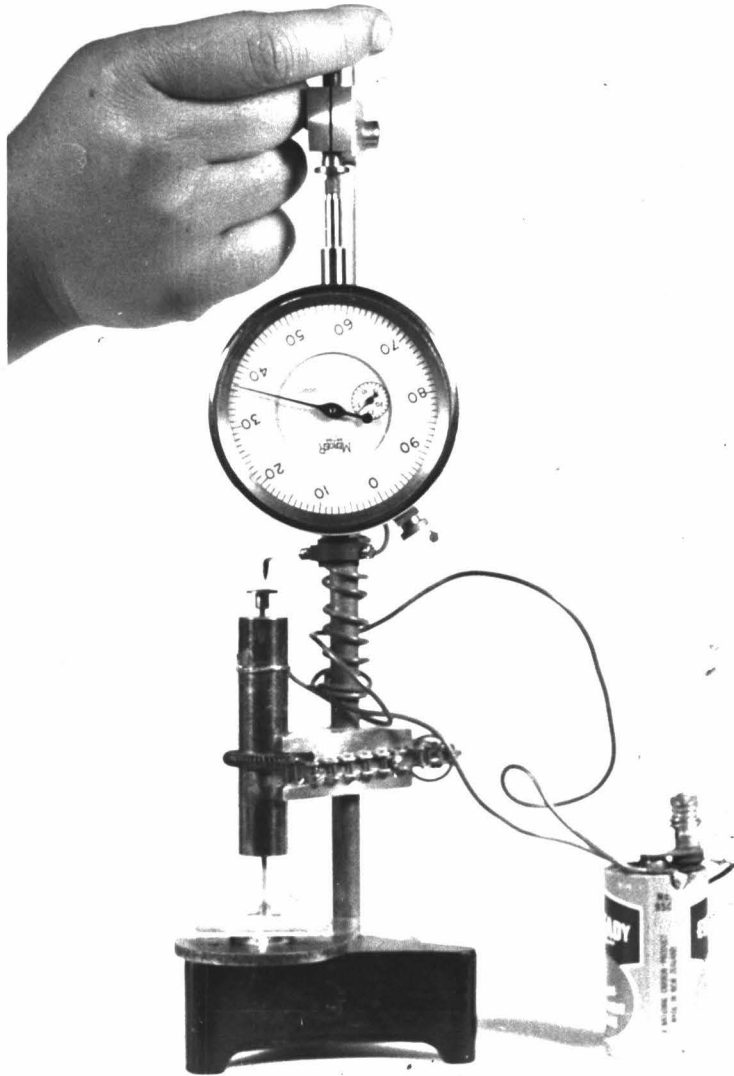
The Instrument consists of a cast base A carrying a rod B on which the various components are mounted. An adjustable block C with a v shaped end carried the guide D for the measuring rod E. This rod has a top member F carrying a small platinum disc and a lower circular member G which is exactly ten millimetres in diameter. The rod has a flattened portion moving in a slot in



Photograph 1: The Instrument.



Photograph 2: The Instrument plus the six millimetre block and the glass slide at the zero position.



Photograph 3: The Instrument plus the three millimetre block, the glass slide and the skin sample being screwed down to obtain the skin thickness measurement.

guide D, thus allowing free vertical movement without permitting rotation. The base casting carries a platen H on which the sample is placed. Measurement of the movement of E is carried out by means of the dial gauge I which is calibrated in $\frac{1}{1000}$ ths of an inch. This gauge is mounted on the immovable block J sliding on rod B to which A is locked by a set screw as in the block C. The lower end of the screwing rod of the dial gauge carries a small insulated bush K to which is attached a feeler arm L with a platinum tip. Around the top end of the dial gauge rod M is another block fitting the stand rod B through which a firm screw adjustment N passes. The platinum contact at the end of feeler arm L and the platinum disc affixed to F complete an electric circuit through the indicator lamp P.

Operation: First the zero reading is obtained on the dial gauge by allowing the measuring rod to rest on the platen H. The adjustant screw N is screwed until the indicator lamp flickers and the reading on the dial gauge is noted. A sample is then placed on the platen after the gauge has been allowed to return to its topmost position. The adjustant screw is again turned until the light flickers and the second reading is noted. The difference is recorded.

In order to extend the range of this instrument two gauge blocks three and six millimetres in thickness are used. The zero reading is taken with one or the other or both of these blocks in place. The movement of the gauge rod is about five millimetres. The weight of the

measuring rod is 5.9935 grams which represents a constant pressure of 0.5994 grams per square millimetre on all samples measured.

(ii) Methods for making frozen sections.

After being measured with the Instrument, the sample was put into either 10% formalin or formol saline or gum arabic for 24 hours, prior to section cutting. The thickness of each section was measured by means of a philomicrometer mounted on a travelling microscope.

(iii) Other instruments and materials.

Following the clipping of wool on the site to be sampled, several types of safety razor and a blade razor were tried to complete wool removal from the skin.

Two instruments similar in principle to a franking stamp, one marking a rectangle of the area of six centimetres (consisting of six two by three one square centimetre squares) and the other a square of four centimetres (consisting of four two by two one square centimetre squares), were used.

Samples were always made slightly bigger than the area marked out for measurement, to allow for the unevenness in thickness towards the edges caused by the cutting out operation.

A local anaesthetic of procaine hydrochloride (1% solution) was used. Approximately five cubic centimetres injected subcutaneously around the area to be removed ensured a painless operation five minutes later.

A scalpel used in conjunction with a pair of forceps, and a pair of shearing blades used on a raised fold of the skin were tried as methods of skin removal.

A Kelvinator refrigerator running at a temperature of four degrees Centigrade and a McAlpine deep freeze running at -15 degrees Centigrade were tried as means for preserving the skin.

(b) Experimental Animals.

Three Romney wether hoggets in store condition were used for the pilot trials and Preliminary Experiment I. Three samples, three by four centimetres per sample, were taken from each animal within a period of four weeks and the skin was taken from positions all over the body of the sheep.

Four samples, two by two centimetres per sample, were taken from the middle of the back of a fourth Romney wether hogget, 16 square centimetres of skin being removed at the one operation.

Throughout the experiments, the animals remained healthy and unaffected by the wounds. The wounds, despite their size and shape healed in a few days by first intention and in no case was trouble experienced through infection. Four weeks after skin removal, the clipped wool and a thin scar were the only indications that the operation had taken place. Pictures of the areas sampled, taken at various times after sampling appear in Chapter III.

II. NATURE AND SCOPE OF EXPERIMENTS.

In this part of the investigations three problems faced the experimenter: firstly, designing instruments and developing techniques which would comply with the

requirements outlined in the Introduction; secondly, acquiring skill in the efficient use of both instruments and techniques; and lastly, defining the errors involved in the techniques and the variations encountered in the experimental material.

A pilot trial showed that safety razors would not remove wool completely even after the sample area had been closely clipped, but that a blade razor was satisfactory. A depilatory was not used because it was thought that it would alter the histological structure and chemical composition of the samples.

A method for the removal of skin from the sheep was tried whereby a fold of skin was picked up and cut off with a pair of shearing blades. However, it was difficult to pick up a large enough skin fold (especially along the back of the sheep) and moreover, a sample thus obtained was uneven in shape and thickness. A scalpel was used to remove skin from the sheep. Frozen sections made from samples removed with a scalpel from live sheep and from pelts removed from sheep at slaughter showed that in both cases the epidermis and the dermis plus some subcutis were removed. The amount of subcutis removed was variable, but did not exceed one to two percent of the total thickness. Since this amount represents in a sample two millimetres in thickness, 0.02 to 0.04 millimetres and since the Instrument readings were taken to only 0.1 millimetre, this error was disregarded. It was

therefore considered/that careful removal of the skin with a scalpel ensured that comparable cell layers were taken off at each sampling and that moreover, these layers correspond with those removed with the pelt at slaughter.

(a) Preliminary Experiment I

(1) Introduction.

This experiment was designed to find out:-

(1) Whether the Instrument gave valid results for the measurement of skin thickness.

A pilot trial showed that the Instrument measurements were highly repeatable whilst examination of the frozen sections showed that the samples presented for measurement represented the same number of cell layers. However, since the skin surface on both sides is undulating, it was not known whether the Instrument measured the "crests", the "troughs" or the means of "crests and troughs", and whether this varied between samples of different thickness. Since the Instrument operates at a constant pressure, it was possible that a thicker sample would be less compressed relatively (though perhaps not absolutely) than a thinner one and that the effect might be one of measuring the "crests", whilst conversely in a thinner sample the "troughs" would be measured. This factor would be important in semi-elastic materials with extreme differences in thickness. It was essential to determine whether this factor was of importance in the range of thicknesses obtained in the skin of sheep.

(2) Whether skin samples could be shaved after removal from the animal.

If samples could be shaved after removal from the animal without altering in skin thickness, it would be

possible to sample unshaved pelts where sheep could not be handled prior to slaughter. Furthermore, sampling would not be restricted to places where hot water, necessary for shaving, was available. The time required for handling the animals during the sampling would also be reduced.

(3) Whether skin samples could be stored for a period of 24 hours without altering in thickness.

If samples could be shaved after removal from the animal and if they could be subsequently stored for 24 hours without altering in thickness, the number of animals which could be sampled per day would be increased.

(2) Plan of Experiment.

A plan of this experiment appears in Table I.

TABLE I

PLAN OF PRELIMINARY EXPERIMENT I

Lot	Piece	Method of Wool Removal	Initial Measurement	Method of Storing	Second Measurement	Soaked for 24 hours prior to making frozen sections	Third Measurement	
PART A	I	1 Shaved on animal	Gauge	Deep freeze	Gauge	Formalin	Philomicro- meter	
		1A Shaved after taking off animal	"	"	"	"	"	
	II	2 Shaved on animal	"	"	"	"	"	
		2A Shaved after taking off animal	"	"	"	"	"	
	III	3 Shaved on animal	"	Refrigerator	"	Formol Saline	"	
		3A Shaved after taking off animal	"	"	"	"	"	
	IV	4 Shaved on animal	"	"	"	"	"	
		4A Shaved after taking off animal	"	"	"	"	"	
	PART B		5 Shaved on animal	"			Gum Arabic	"
			6 Shaved on animal	"			"	"

The term "piece" refers to a rectangle containing six equal squares (two by three). A "lot" consists of two adjacent "pieces", being a rectangle containing 12 squares (three by four). Throughout the thesis the term "square" refers to a square of skin with an area of one square centimetre, i.e. the sides of a square are equivalent to the diameter of that part of the Instrument which comes in direct contact with the skin sample. Each square was measured five times with the Instrument and the mean was calculated. Three frozen sections were taken at random from each square and measured by the philomicrometer method. Each section was measured in ten places and the mean of the 30 measurements obtained.

In Part A of this experiment, four lots of skin were removed at four samplings. One piece in each lot was shaved while still on the animal and the other after it had been taken off. After being measured with the Instrument, two lots (four pieces) were put into a deep freeze and two lots into a refrigerator for 24 hours, and then measured again with the Instrument, care being taken to defrost the pieces which had been in the deep freeze. After this second measurement, two lots were put into formalin and two lots into formol saline for 24 hours, prior to the making of frozen sections which were measured by means of the philomicrometer mounted on a travelling microscope.

In Part B, two pieces (non-adjacent) of different thicknesses were shaved whilst still on the animal, measured with the Instrument and put into gum arabic. After 24 hours in the latter, frozen sections were made, measured as in Part A of this experiment with a philomicrometer and the two measurements compared.

The sampling positions have already been discussed under Section I (b) of this chapter.

(3) Results.

Mean thicknesses of skin samples shaved before and after removal from the animal are presented in Table 2.

The significance of the differences between these means was tested by an Analysis of Variance using the method of weighted means (Snedecor, 1946).

A close similarity of the means and the non significant "F" ratio "between treatments" indicate that skin may be shaved after removal from the animal without appreciable alteration in thickness.

Table 3 shows that there was a difference, significant at the 1% level, "between lots". This is not surprising because each lot was taken from a different part of the body of the animal and subsequent experiments (see Chapter II) showed a wide variation in skin thickness over the body of the sheep. The "treatment x piece interaction" was non significant. This means that all the pieces measured responded similarly to the treatment in spite of the wide variation in thickness between pieces. In other words neither the thinner nor the thicker pieces altered differentially in thickness as a result of

being shaved after removal from the animal. The differences "between squares within treatments" were significant at the 1% level. This means that the variation in skin thickness within a sample was much greater than the variation between repeat measurements of the same sample. The Instrument was shown therefore, to have sufficient accuracy for the measurement of skin thickness, taking into account the magnitude of the variations occurring within the experimental material.

All the Analysis of Variance calculations in Chapter IA are, for convenience, in tenths of a millimetre.

TABLE 2.

THE EFFECT ON SKIN THICKNESS OF SHAVING BEFORE
AND AFTER REMOVAL OF THE SAMPLE FROM THE
ANIMAL

Lot	Shaving Before		Shaving After	
	Piece	Mean (mm)	Piece	Mean (mm)
I	1	2.5	1A	2.5
II	2	3.0	2A	2.9
III	3	3.0	3A	3.1
IV	4	2.2	4A	2.2
		2.7		2.7

TABLE 3.

ANALYSIS OF VARIANCE OF THE SAME DATA AS
IN TABLE 2.

(Instrument Measurements)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between treatments	1	0.01	< 1	N.S.
Between lots	3	917.99	97.46	* *
Treatments x pieces	3	9.42	1.55	N.S.
Between squares within treatments	40	6.08	28.95	* *
Between measurements within squares	192	0.21		
Total	239			

N.S. = non significant
* = significant at the 5% level.
** = significant at the 1% level.

Mean thicknesses of skin samples measured immediately after removal from the animal and after 24 hours in the deep freeze are presented in Table 4. After storage in the deep freeze an average increase in the means of 7.5% was observed and Table 5 shows "between treatments" differences to be significant at the 5% level. Therefore, in terms of the requirements outlined in the Introduction, this method is not satisfactory for the preservation of samples prior to the measurement of skin thickness.

Mean thicknesses of skin samples measured immediately after removal from the animal and after 24 hours in the refrigerator are presented in Table 6. A close similarity of the means and the non-significant "between treatments" "F" ratio shown in Table 7 indicate that skin may be stored for 24 hours in a refrigerator without appreciable alteration in skin thickness.

Tables 5 and 7 show that there was a difference significant at the 1% level "between pieces". This is to be expected because each piece was taken from a different part of the body of the animal and subsequent experiments (see Chapter II) showed a wide variation in skin thickness over the body of a sheep. The magnitude of the "treatments by pieces", the "between squares within treatments" and the "between measurements within squares" mean squares was of the same order as shown in Table 3 and the same comments apply.

TABLE 4

THE EFFECT ON SKIN THICKNESS OF STORAGE FOR

24 HOURS IN A DEEP FREEZE

(Instrument Measurements)

Lot	Fresh Piece	Fresh Mean (mm)	After deep freeze Mean (mm)	Increase	
				mm.	%
I	1	2.5	2.8	0.3	12.0
I	1A	2.5	2.6	0.1	4.0
II	2	2.9	3.1	0.2	6.9
II	2A	2.9	3.1	0.2	6.9
	Mean				7.5

TABLE 5

ANALYSIS OF VARIANCE OF THE SAME DATA AS

IN TABLE 4

(Instrument Measurements)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between treatments	1	141.06	14.42	*
Between pieces	3	342.95	35.06	* *
Treatments x pieces	3	9.78	5.34	N.S.
Between squares within treatments	40	1.83	7.32	* *
Between measurements within squares	192	0.25		
Total	239			

TABLE 6

THE EFFECT ON SKIN THICKNESS OF STORAGE FOR
24 HOURS IN A REFRIGERATOR
(Instrument Measurements)

Lot	Fresh		After refrigerator	
	Piece	Mean (mm)	Mean (mm)	
III	3	3.0	3.0	
III	3A	3.1	3.2	
IV	4	2.2	2.2	
IV	4A	2.2	2.2	
	Mean	2.6	2.6	

TABLE 7

ANALYSIS OF VARIANCE OF THE SAME DATA AS IN

TABLE 6
(Instrument Measurements)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between treatments	1	0.15	< 1	N.S.
Between pieces	3	1,526.30	1,838.90	* *
Treatments x pieces	3	0.83	< 1	N.S.
Between squares within treatments	40	8.24	103.00	* *
Between measurements within squares	192	0.08		
Total	239			

In Table 8 mean thicknesses of skin samples measured with the Instrument immediately after removal from the animal are compared with the philomicrometer measurements of frozen sections previously soaked for 24 hours in 10% formalin. Since the frozen sections showed an average increase in thickness of almost 25% this method was abandoned.

In Table 9 mean thicknesses of skin samples measured with the Instrument immediately after removal from the animal are compared with the philomicrometer measurements of frozen sections previously soaked for 24 hours in formol saline. Since the frozen sections showed an average increase in thickness of almost 34%, this method was also discontinued.

In Table 10 mean thicknesses of skin samples measured with the Instrument immediately after removal from the animal are compared with the philomicrometer measurements of frozen sections previously soaked for 24 hours in gum arabic. A close similarity was observed between the means, a correlation of 0.9895 being obtained between the Instrument and the philomicrometer measurements. A similar correlation was obtained for both the thinner and the thicker samples. Therefore, in terms of the requirements outlined in Section II (a) 1 of this chapter, the Instrument was shown to be satisfactory and the time consuming frozen section method of measurement of skin thickness was abandoned.

TABLE 8

COMPARISON OF THE MEASUREMENTS OF SKIN THICKNESS
OBTAINED BY INSTRUMENT MEASUREMENTS OF FRESH TISSUE
WITH PHILOMICROMETER MEASUREMENTS OF FROZEN
SECTIONS PREVIOUSLY KEPT FOR 24 HOURS IN FORMALIN.

Piece	No. Square	Mean Measurements (mm)		Increase	
		Instrument	Philo- micrometer	m.m.	%
1	1	2.8	3.5	0.7	25.0
	2	2.8	3.5	0.7	25.0
	3	2.8	3.4	0.6	21.4
1A	1	2.8	3.6	0.8	28.6
	2	2.6	3.3	0.7	26.9
	3	2.4	2.9	0.5	20.8
Mean				0.7	24.6

TABLE 9

SIMILAR COMPARISON AS IN TABLE 8, BUT SAMPLES
KEPT FOR 24 HOURS IN FORMOL SALINE

Piece	No. Square	Mean Measurements (m.m.)		Increase	
		Instrument	Philo- micrometer	m.m.	%
3	1	3.0	4.2	1.2	40.0
	2	3.0	3.8	0.8	26.7
3A	3	3.2	4.4	1.2	37.5
4	1	3.1	3.8	0.7	22.6
4A	1	3.1	4.3	1.2	38.7
	2	3.1	4.3	1.2	38.7
Mean					34.0

TABLE 10

SIMILAR COMPARISON AS IN TABLE 8, BUT SAMPLES
KEPT FOR 24 HOURS IN GUM ARABIC.

No. Piece	Square	Mean Measurements (mm)		Difference (m.m.) (Approx.)
		Instrument	Philo- micrometer	
5	1	2.1	2.2	+ 0.1
	2	2.0	2.1	+0.1
	3	2.2	2.2	
	4	2.1	2.2	+ 0.1
	5	2.2	2.3	0.1
	6	2.2	2.2	
6	1	3.0	3.1	+ 0.2
	2	2.7	2.8	+ 0.1
	3	2.8	2.8	
	4	2.9	3.0	+ 0.1
	5	2.6	2.6	
	6	2.7	2.7	
Mean		2.45	2.5	0.1

(b) Preliminary Experiment II.

(1) Introduction.

As a result of Preliminary Experiment I, the Instrument was accepted as an accurate method for obtaining skin thickness measurements. Since the shaving of skin following its removal from the animal and the keeping of the sample in a refrigerator for 24 hours prior to measurement appeared to be the best procedure for use in subsequent experiments, it was thought advisable to confirm its validity. It was also realized that a uniform sampling area must be obtained. The gradients in skin thickness over the

body of the animal were mapped out (see Chapter II) and samples were taken within an area uniform in thickness. This experiment was designed therefore, to confirm the suitability and to define the errors of the technique to be used in future experiments, and the variations encountered in the experimental material.

(2) Plan of Experiment.

As shown in Diagram 1, four samples were taken from the middle of the back of a sheep. Pieces (1) and (3) were adjacent samples situated an inch to the left of the midline and half way between the shoulder and the hip bone. Pieces (2) and (4) were adjacent samples similarly situated to pieces (1) and (3) but on the right side of the animal.

Pieces (1) and (2) were shaved before removal from the animal while pieces (3) and (4) were shaved afterwards. All four pieces were measured both immediately after removal from the sheep and after being kept for 24 hours in a refrigerator.

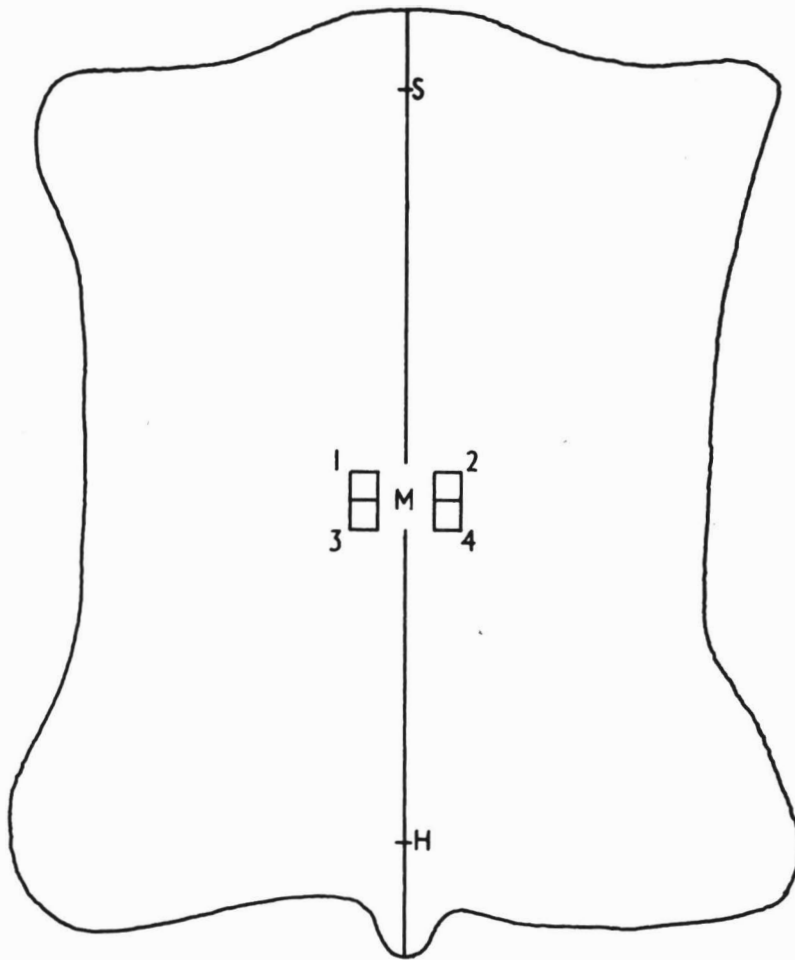
The term "piece" refers to a square of skin with an area of four square centimetres, divided into four equal squares. A "square" has been defined in Section II (a) (2). Each square was measured five times with the Instrument and the mean was obtained.

(3) Results.

Mean thicknesses of skin samples shaved before and after removal from the animal are presented in Table 11. A close similarity of the means and the non significant "between treatments" "F" ratio as shown in Table 12, confirm the validity of shaving

DIAGRAM I

Sampling positions used in the preliminary experiment II



S Vertebral column at the shoulder level.

H Vertebral column at the hip bone level.

SM=HM

1 & 3 Adjacent pieces an inch to the left of the vertebral column and half way between the shoulder and the hip bone.

2 & 4 Adjacent pieces an inch to the right of the vertebral column and half way between the shoulder and the hip bone.

the skin after removal from the animal.

Mean thicknesses of skin samples measured immediately after removal from the animal and after 24 hours in the refrigerator are presented in Table 13. A close similarity of the means and the non significant "between treatments" "F" ratio shown in Table 14 confirm the suitability of using the refrigerator for skin storage.

Tables 12 and 14 show that due to the selection of a sampling area uniform in thickness, the variation "between pieces" was reduced in comparison with the values obtained in Preliminary Experiment I. The "between measurements within squares" mean square was much smaller than either the "between squares within pieces" or the "between pieces within treatment" mean squares. This means that most of the variance was due to the variability in thickness within and between samples. Numerous repeat measurements of the same square with the Instrument are therefore unnecessary.

Estimates of the components of variance appear in Table 15. The "between pieces within treatments" component of variance, i.e. due to the variations in thickness between samples taken from a uniform area, was 0.005 millimetre. The "between squares within pieces" component of variance, i.e. due to variations in thickness within a piece or sample, was 0.002 millimetre. The "between measurements within squares" component of variance, i.e. due to variations between the repeat Instrument measurements of the same square, was 0.002 millimetre. The total of the variance

components was 0.009 millimetre.

TABLE 11.

THE EFFECT ON SKIN THICKNESS OF SHAVING BEFORE
AND AFTER REMOVAL OF THE SAMPLE FROM THE ANIMAL.
(Instrument Measurements)

Shaven Before		Shaven After	
Piece	Mean (mm)	Piece	Mean (mm)
1	1.8	3	1.9
2	1.9	4	2.0
Mean	1.8		1.9

TABLE 12.

ANALYSIS OF VARIANCE OF THE SAME DATA AS IN

TABLE 11. (Instrument Measurements)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between treatments	1	4.05	< 1	N.S.
Between pieces within treatments	2	11.02	9.84	N.S.
Between squares within pieces	12	1.12	5.33	*
Between measurements within squares	64	0.21		
Total	79			

TABLE 13.

THE EFFECT ON SKIN THICKNESS OF STORAGE FOR 24
HOURS IN A REFRIGERATOR.
(Instrument Measurements)

Fresh		After refrigerator	
Piece	Mean (mm)	Mean (mm)	
1	1.8	1.9	
2	1.9	1.9	
3	1.9	1.8	
4	2.0	2.0	
Mean	1.9	1.9	

TABLE 14.

ANALYSIS OF VARIANCE OF THE SAME DATA AS IN

TABLE 13.

(Instrument Measurements)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between treatments	1	0.63	< 1	N.S.
Between pieces within treatments	6	8.66	3.90	N.S.
Between squares within pieces	24	2.22	11.68	* *
Between measurements within squares	128	0.19		
Total	159			

TABLE 15.

INSTRUMENT MEASUREMENTS OF SKIN THICKNESS.

ESTIMATES OF THE COMPONENTS OF VARIANCE.

Source of Variation	M.S.	Individuals per subclass	M.S. is an estimate of	Variance Component (mm)
Between pieces within treatments	11.02	20	$6_M^2 + 56_S^2 + 206_P^2$	$6_P^2 = 0.005$
Between squares within pieces	1.12	5	$6_M^2 + 56_S^2$	$6_S^2 = 0.002$
Between measurements within squares	0.21	1	6_M^2	$6_M^2 = 0.002$

(4) Sampling procedure finally adopted.

The animal was caught, its legs were tied together and it was laid on its side. The wool on the sample site was clipped and local anaesthetic was injected subcutaneously. If wool was to be removed while the skin was still on the animal, the area was

wetted and soaped, and the wool shaved. The square or rectangle to be removed was marked out and by this time the anaesthetic was taking effect. The skin was removed with a scalpel and put into a tightly stoppered jar. Some sulphanilamide was put on the wound and the animal was released. If not shaved previously, the sample was then shaved and trimmed. The blood was washed off and the skin blotted on filter paper to remove surplus moisture, before being replaced in the jar. After measurement with the Instrument, the sample was put into a refrigerator for 24 hours and measured again. The whole process took about ten minutes per sheep. The instruments used are shown in photograph 4. Photographs 5 to 15 inclusively, illustrate the sampling procedure.

DISCUSSION.

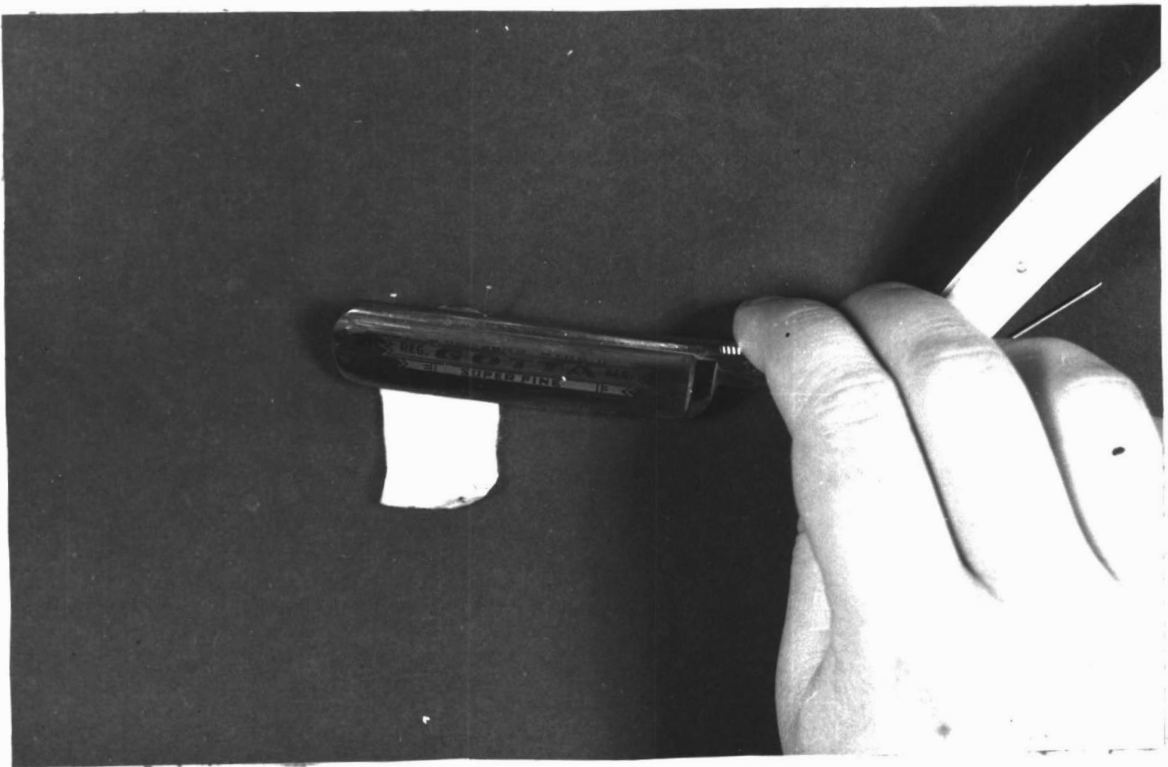
The Instrument used in these experiments is accurate, quick and easy to use. It is useful for experiments where part of the skin sample is used for histological sections and part for chemical analyses, since both of these require the removal of the skin from the animal. Also for chemical analysis a comparatively large sample is necessary. For the measurement of skin thickness with the Instrument, over one square centimetre of skin has to be removed from the animal. Both the experiment described in this Chapter and those of Chapters III and IV, indicate that the health of the animals



Photograph 4: The equipment used for the skin removing operations in the field:-
(left to right - back row)
the sterilizer, the marking apparatus, the air-tight jars for the skin samples.
(left to right - front row)
the container for hot water, the blade razor, the scalpel, the forceps, the scissors, the soap and the sulphanilamide powder.



Photograph 5: Shaving before skin removal from a sheep.



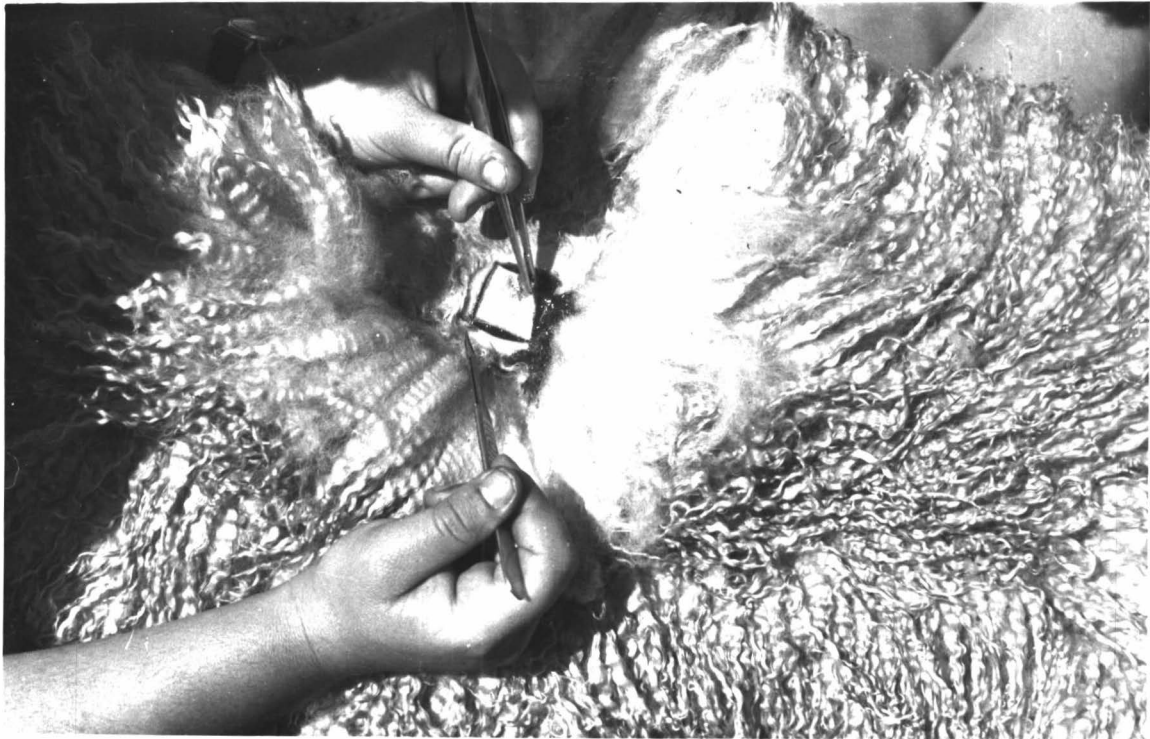
Photograph 6: Shaving after skin removal from a sheep.



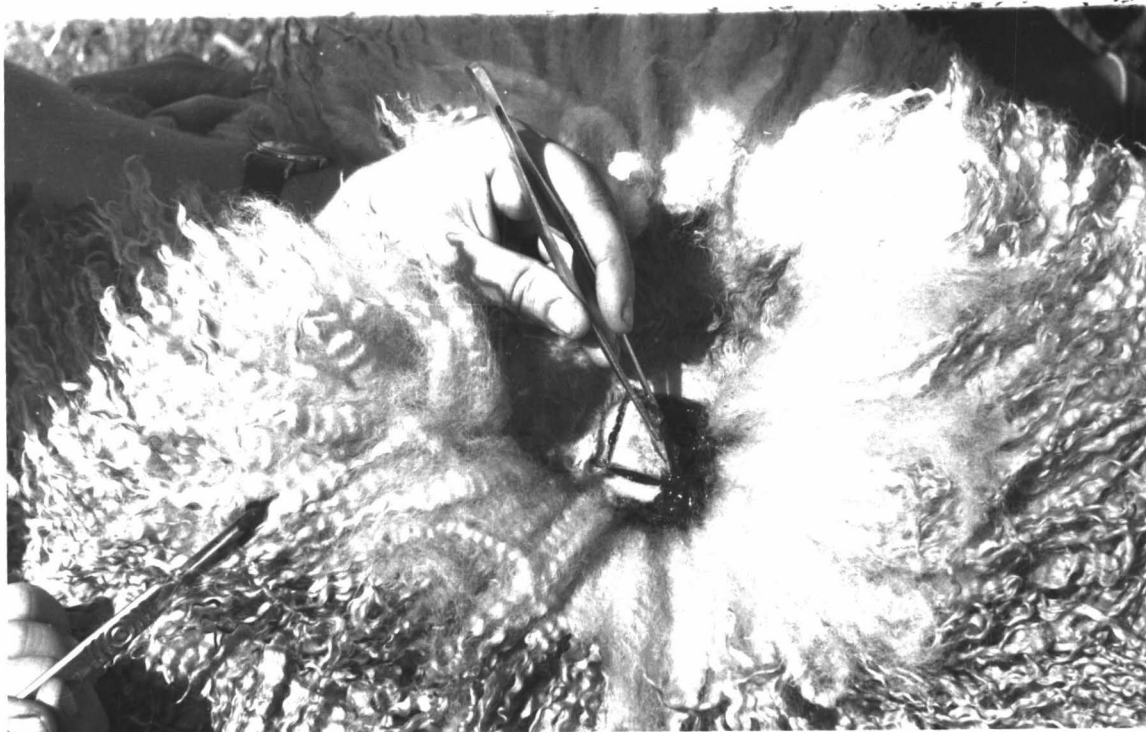
Photograph 7: Marking of the area of skin to be removed.



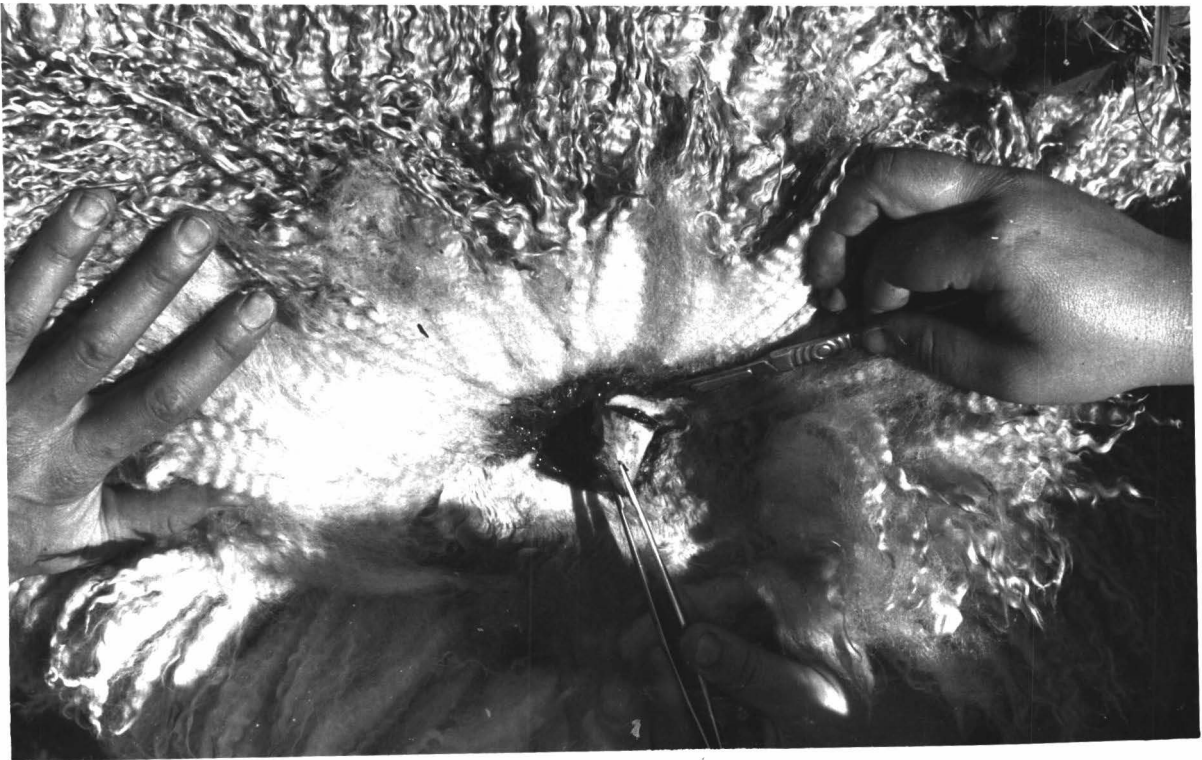
Photograph 8: The marked area of skin before removal from the sheep.



Photographs 9 and 10: The progress of the skin removal operation.



Photographs 11 and 12: The progress of the skin removal operation.



Photograph 13: The progress of the skin removing operation.



Photograph 14: The progress of the skin removing operation, all of the skin sample except one side has been detached from the body of the sheep.



Photograph 15: Skin has been removed from the sheep and the wound is dusted over with sulphamide powder.

appears to be unaffected even by the removal of three samples of 12 square centimetres each within four weeks from adult sheep, or monthly sampling of four square centimetres from lambs. However, for large scale work, this method is disadvantageous because there is some reduction in both pelt and fleece values and because the number of samplings must be restricted both in number and frequency.

Where skin thickness values alone are required, a "pinch" type of instrument similar to those used by Nicov (1931) and Carstens and Kinzelbach (1933), should it prove to be an accurate method for the measurement of skin thickness, would be preferable, since skin would not have to be removed from the animal for the measurement. Even where changes in cell structure of the skin are being investigated as well, the "pinch" method would be useful because the amount of skin required for histological sections is only of the order of a few square millimetres. Thus the use of this method would make possible the following of thickness and histological changes in the skin of the same animal at frequent intervals.

The technique for measurement of skin thickness, described in this chapter, could also be used, in future work, to test the validity of other instruments used for the same purpose.

SUMMARY

(1) The Instrument is a quick and accurate method for measurement of skin thickness.

(2) Careful removal of the skin from the animal by means of a scalpel, results in uniform samples and the number of cell layers removed, corresponds with that removed with the pelt at slaughter.

(3) The blade razor used while the skin is on the animal or after it has been taken off, is the best method for wool removal without altering the nature of the skin itself.

(4) Skin can be preserved in a refrigerator for 24 hours without altering in thickness.

B. CHEMICAL ANALYSIS.

INTRODUCTION.

Before fat and protein variations in the skin of growing and adult sheep could be examined, the ether extraction and Kjeldahl techniques had to be modified. It was also essential to define the errors involved in the chemical analyses and the variations occurring in the experimental material. Experiments were designed to determine:-

- (a) the effect on chemical composition of any water loss due to the drying out of skin between sampling and weighing;
- (b) the length of time required for complete fat extraction;
- (c) the extent of contamination of fat extracts with protein;
- (d) the magnitude of errors due to methods of determination of fat and protein;
- (e) the size of variations in chemical composition between samples taken from different positions on a sheep;
- (f) the magnitude of day to day fluctuations in the chemical composition of the skin in sheep.

No measure could be obtained of the possible water imbibition or loss during the washing of the sample to remove blood and its subsequent drying with a filter paper to remove excess moisture. However, since all samples were treated alike, it was considered that these errors would be almost constant and could be ignored in a study concerned with relative rather

than absolute values.

I. METHODS AND MATERIALS.

(a) Apparatus

The apparatus is shown in Photographs 16, 17 and 18. It consists of:-

(1) Extractor with the following dimensions:

Length of barrel: 4.3 - 5.0 centimetres

Internal diameter of
barrel: 1.25 centimetres

Internal diameter of
siphon tube: 2 millimetres

To prevent the carrying over of small pieces of solid material, the siphon tube is plugged with cotton wool at its point of junction with the barrel. The lugs near the top of the barrel enable it to hang freely within the collecting tube. Before use, the extractor is cleaned, dried overnight in an oven at 85 degrees Centigrade, plugged with dry cotton wool, cooled in a desiccator and weighed.

(ii) Collecting tube:

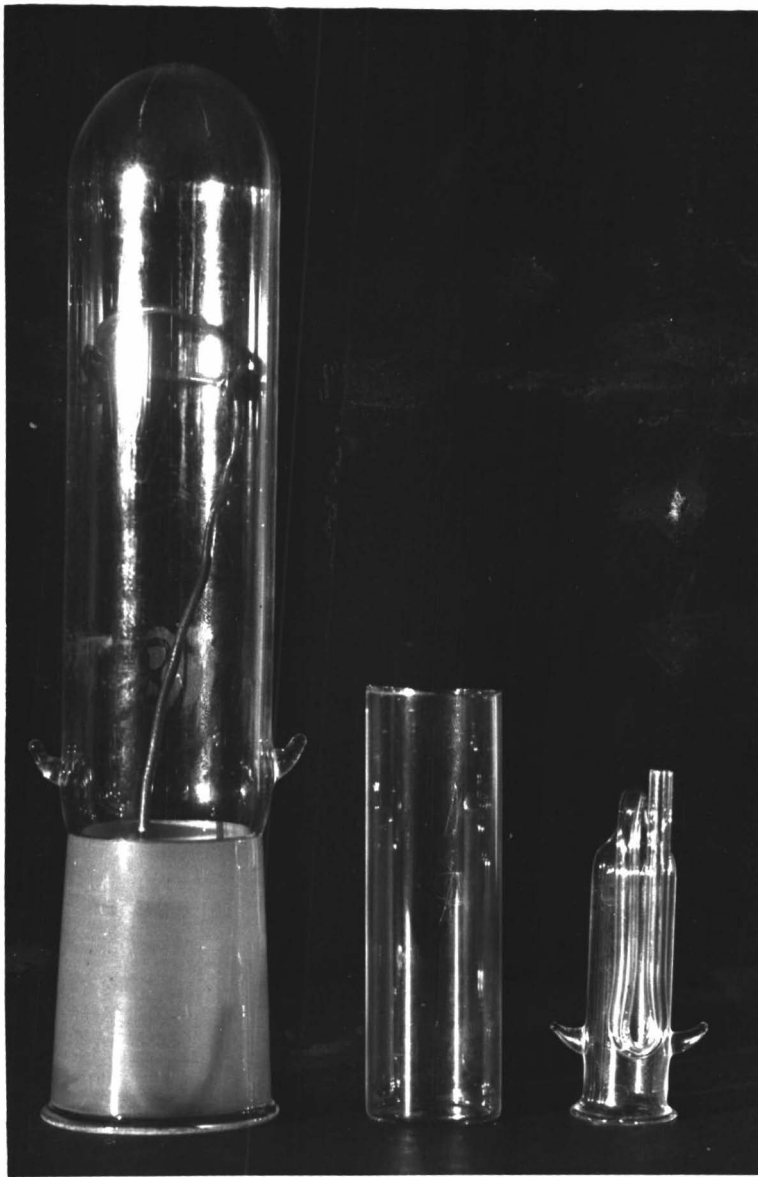
This consists of a five by one inch flat-bottomed sample tube cut down to a height of from 3 to 3.5 inches. To the cleaned, dried and cooled tube are added before weighing and use numerous pieces of very finely divided porous pot to prevent bumping during the ether extraction.

(iii) Outer Jacket with the following dimensions:

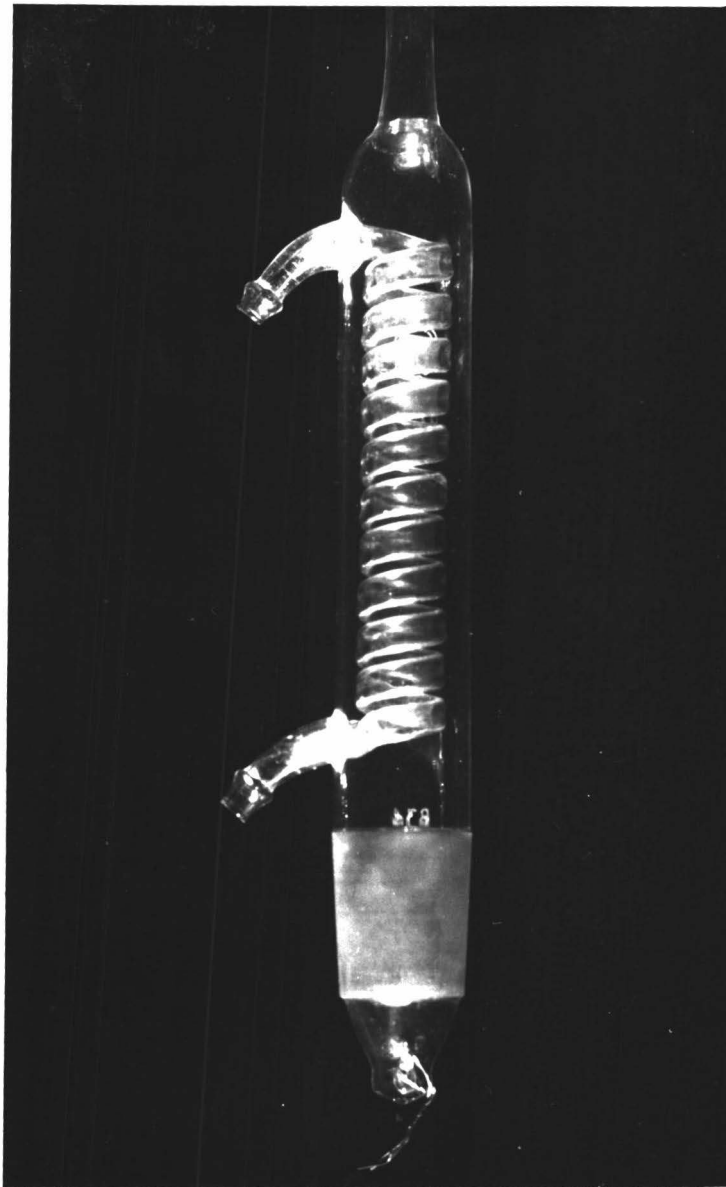
Total length: 7 - 7.5 inches

Internal diameter: 1.5 inches

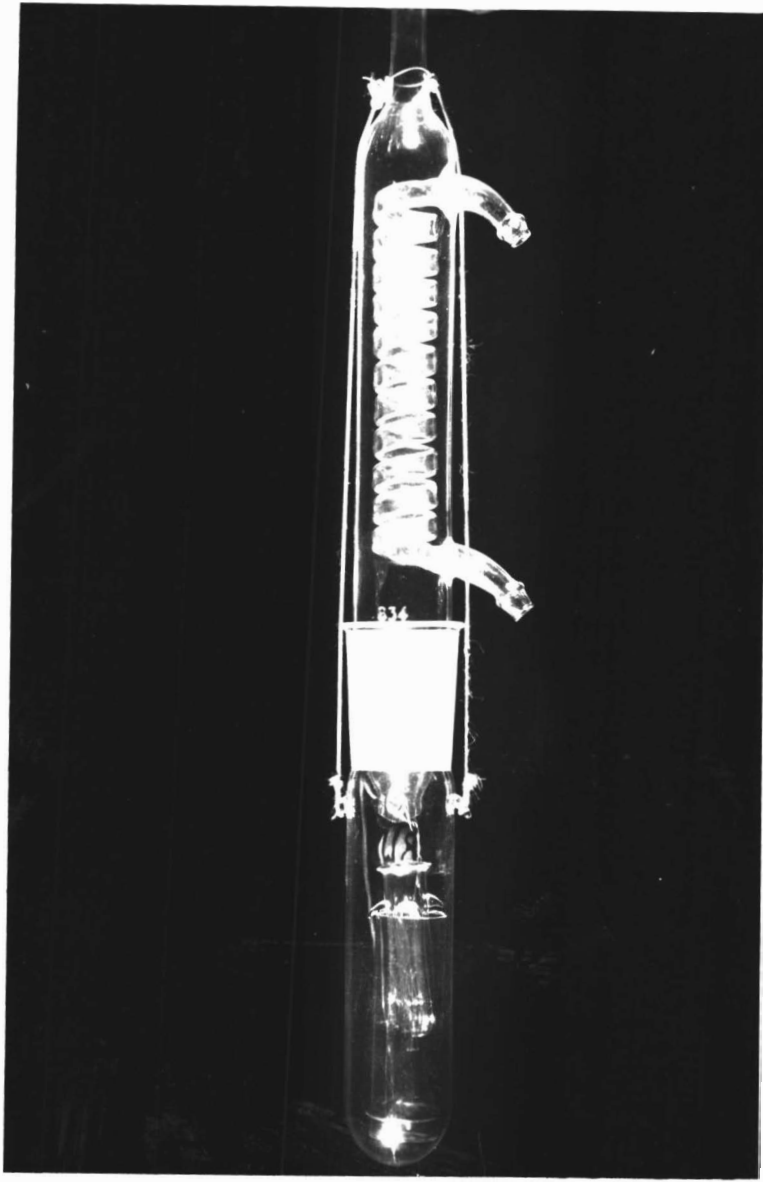
Size of standard
joint socket at top: B 34



Photograph 16: (Left to right) The bottom part of the condenser, the tube and the extractor.



Photograph 17: The top part of the condenser.



Photograph 13: The extractor and tube inside the condenser prior to being placed over a water bath and connected to running water for the ether extraction process.

The lugs just below the socket enable it to be held tightly to the condenser.

Standard Kjeldahl apparatus was used for protein estimation.

(b) Sampling Procedure and Analysis.

After removal from the animal, the skin sample was cleaned of subcutaneous tissue and the blood was washed off with water. The sample was dried with filter paper and placed in a tightly stoppered jar. After a time varying from one to three hours, it was put into a refrigerator for 24 hours. Since samples were removed from the sheep at the woolshed, about a mile from the refrigerator, and six to 16 animals were sampled at a time, this large variation in time between sampling and weighing was unavoidable. After 24 hours in a refrigerator, the skin was measured with the Instrument, the time of exposure to air being three to five minutes.

It was then replaced in the jar, weighed (the jar was weighed later when dry and empty to obtain the weight of the sample by difference) and covered with absolute alcohol. Samples used for chemical analysis were approximately four square centimetres (two by two centimetres) in area and two to three grams in weight. After 24 hours, the alcohol was poured into an evaporating dish and evaporated in an oven at 85 degrees Centigrade. The skin was again soaked in absolute alcohol for at least six hours, the extract being then drained

into the same dish, similarly evaporated and dried in the oven for 24 hours. The skin together with the scrapings from the evaporating dish and the jar was then put into the dried and weighed extractor and extracted with ether for 48 hours, in the apparatus described. Ether was then evaporated by detaching the condenser. The extractor and tube were dried for 24 hours in an oven at 85 degrees Centigrade, cooled in a desiccator and weighed separately.

Then if F_1 = weight of the tube plus fat
 F_2 = weight of the tube
 P_1 = weight of the extractor plus
the fat-free residue
 P_2 = weight of the extractor
then $F_1 - F_2$ = fat contained in the sample
 $P_1 - P_2$ = fat-free residue contained in
the sample
and $(F_1 - F_2) + (P_1 - P_2)$ = dry matter contained in
the sample.

From the skin residue, cut up to enable random sampling, two 0.06 - 0.08 gram portions were weighed for nitrogen estimation. This was carried out by the Kjeldahl method, using for the digestion a 50:1 potassium sulphate - selenium catalyst mixture and a time of one-and-a-quarter hours. One-twentieth of the diluted digest was taken for micro-distillation, the ammonia evolved being trapped in boric acid solution and titrated with 0.01 N potassium bi-iodate with bromocresol green-methyl red indicator. Protein was calculated by multiplying the nitrogen content thus obtained by the factor 6.25.

The similarity of values for the protein content of skin, obtained by taking the weight of the dried, fat free residue and by the Kjeldahl method was observed. On the results obtained in this chapter it seemed that within the limits of experimental error, the fat free residue was entirely composed of protein.

II. NATURE AND SCOPE OF EXPERIMENT.

- (a) The effect on chemical composition of any water loss due to the drying out of skin between sampling and weighing and
- (f) The magnitude of day to day fluctuations in chemical composition of the skin of sheep.

To determine the extent of the water losses between sampling and weighing, and also the day to day fluctuations in chemical composition, a sample was taken from each of three hoggets on each of two consecutive days and weighed:-

- (i) immediately after the removal of adhering subcutaneous tissue and blood;
- (ii) after one hour in a tightly stoppered jar standing (at air temperature);
- (iii) after three hours in a tightly stoppered jar standing at air temperature;
- (iv) after four hours in a tightly stoppered jar standing at air temperature;
- (v) after 24 hours in a tightly stoppered jar standing in a refrigerator;
- (vi) after five minutes' exposure to air.

These treatments and weighing times were chosen because they corresponded with those occurring during the routine sampling procedure and analysis. Weighing (i) represented the "true" original weight of the sample except for the possible variation between samples due to the water imbibition or loss during the cleaning up process. However, as suggested in the Introduction, since all samples were treated alike, it was considered that these errors would be almost constant between samples and could be disregarded in a study concerned with relative rather than absolute values. Weighing (ii), (iii) and (iv) represented the variation in time during which the samples were kept under those conditions between removal from the animal and placement in the refrigerator. For example, in the Shearing Experiment (see Chapter IV), the first sample would remain in a tightly stoppered jar for three to four hours and the last one, for less than one hour. It was considered important to determine whether these variations in the length of time a sample was kept in the jar had an appreciable effect on the weights of skin samples and therefore on the calculated fat and protein content of the skin. Weighing (v) was taken because all samples were put into a refrigerator for 24 hours prior to the measurement of skin thickness and it was considered necessary to find the effect of this treatment on the calculated chemical composition. Weighing (vi) represented the normal weighing after measurement of the skin thickness, henceforth referred to as the

"original weight" and used for the calculation of fat and protein contents of the skin.

Results.

Fat and protein contents expressed as percentages of skin samples weighed at weighings (i), (ii), (iii), (iv), (v) and (vi), are presented in Table 16. The effect on the chemical composition, of water losses due to the drying out of skin between sampling and weighing was shown to be negligible being smaller than the errors in the chemical technique and the positional and day to day variations in the chemical composition (see Tables 17 and 20).

TABLE 16

DRYING OUT BETWEEN SAMPLING AND WEIGHING

(Fat and protein content expressed as % of the original weight)

Samples	1	1A	2	2A	3	3A	1	1A	2	2A	3	3A
0 hrs. after sampling	3.3	3.8	3.7	4.5	1.6	1.6	15.4	16.6	14.2	14.3	16.4	15.4
1 " after sampling	3.3	3.8	3.7	4.5	1.7	1.6	15.7	16.8	14.3	14.4	16.6	15.5
3 " after sampling	3.3	3.8	3.7	4.6	1.7	1.7	15.7	16.8	14.3	14.5	16.6	15.8
4 " after sampling	3.3	3.8	3.7	4.6	1.7	1.7	15.8	16.9	14.3	14.5	16.7	15.8
after 24 hrs. in refrig.	3.3	3.8	3.7	4.6	1.7	1.7	15.8	16.8	14.3	14.5	16.7	15.9
after 5 mins. in air	3.4	3.9	3.8	4.6	1.7	1.7	15.8	17.5	14.7	14.6	16.7	16.2

The fat and protein contents of samples, expressed as percentage of the original material and taken from each of three sheep on each of two consecutive days, are presented in

in Table 17. It would be difficult to determine how much of the difference in the fat and protein contents of the skin on the two days, was due to day to day fluctuations and how much was contributed by positional variations and by errors in the techniques of chemical analysis, because samples taken on different days came from different positions on the sheep and included the chemical analysis errors. However, the fact that positional variations exceed differences between samples obtained on two consecutive days suggests that the day to day variations in fat and protein content are relatively unimportant. On the other hand, the samples were taken on two consecutive days only, whereas it is possible that sudden changes in the environmental factors such as temperature, humidity etc. might have caused more marked fluctuations during another period.

TABLE 17

DAY TO DAY VARIATIONS ON THE SAME
SHEEP

(Fat and protein content expressed as % of the original weight)

Sheep	Fat		Protein	
	1st Day	2nd Day	1st Day	2nd Day
1	3.4	3.9	15.8	17.5
2	3.8	4.6	14.7	14.6
3	1.7	1.7	16.7	16.2

(b) The length of time required for complete fat extraction.

To determine the length of time necessary for complete fat extraction, six samples were extracted for 24, 48 and 72 hours and at each stage fat percentage was determined.

Results.

The fat content values for the six samples, expressed as percentage of the original weight, after the different periods of extraction, are given in Table 18. Further fat extraction occurred in only two samples out of six and the extra amount obtained was less than the difference ascribed to errors in the chemical technique (see Table 20). These results therefore indicate that a period of 24 hours is sufficient for complete fat extraction. A period of 48 hours was henceforth adopted to ensure that all fat was extracted, thus allowing for a wide variation in the rate of extraction.

TABLE 18

LENGTH OF TIME REQUIRED FOR COMPLETE FAT EXTRACTION

Sample	Fat content expressed as % of the original weight		
	After 24 hours	After 48 hrs.	After 72 hrs.
1	1.4	1.5	1.5
2	2.4	2.4	2.4
3	2.2	2.2	2.2
4	1.9	1.9	1.9
5	1.9	1.9	1.9
6	1.7	1.6	1.6

(c) Contamination of fat extracts with protein.

Extracted fat was found generally to contain small amounts of nitrogen. To determine whether this arose from entrained protein, six fat samples suspected of being contaminated with protein through accidental seepage of water into the extractors were dissolved in petroleum ether and the solutions were twice extracted with half-normal sulphuric acid. Nitrogen was estimated by the Kjeldahl method.

Then if (i) is the petroleum ether solution after extraction (non-protein nitrogen);

(ii) is the acid solution (acid-soluble non-protein nitrogen plus protein);

and (iii) is the acid solution after treatment with $\text{Ba}(\text{OH})_2$ and ZnSO_4 and filtration (non-protein nitrogen)

(iii) minus (ii) gives the protein content of the fat.

Results.

The nitrogen content of six fat samples, expressed as percentage of the fat sample, are presented in Table 19. In four samples no protein nitrogen was detected, in the remaining two the amount was less than 0.5% of the fat sample. It was concluded that the protein contamination of fat samples was negligible.

TABLE 19

NITROGEN CONTENT OF FAT SAMPLES

Sample	Total N as % of fat sample	Protein N as % of fat sample
1	0.61	0.00
2	0.30	0.00
3	0.29	0.10
4	0.81	0.35
5	0.37	0.00
6	0.51	0.00

- (d) The magnitude of errors due to methods of determination of fat and protein and
- (e) The size of variations in chemical composition between samples taken from different positions on a sheep.

To determine variations in chemical composition between samples taken from different positions on a sheep, one hogget was killed and sampled in six places, which were identical in position with those used in the Skin Development Experiment (Chapter III). Thus sample A (see Table 20) was situated immediately posterior to the shoulder, sample C immediately anterior to the hip bone and sample B half-way between A and C. A, B and C were located an inch to the left of the vertebral column. Samples D, E and F came from similar positions as samples A, B and C respectively but on the right side of the animal.

Each sample was divided into two and the fat and protein content of each half were determined to obtain a measure of the errors in the chemical analysis.

Large samples (two by four centimetres) were taken from each position because division of the usual samples (two by two centimetres) would have produced disproportionately large weighing errors. The suffix ' is used to distinguish between the two halves (henceforth called duplicates) of the skin sample removed from a given position.

Results.

The chemical composition of samples removed from the different positions and of the duplicates, is shown in Table 20. Table 21 indicates that the differences in fat content between positions were bigger than the ones within positions or between duplicates, and that this difference is significant at the 1% level. Table 22 shows that the "between positions" component of variance was 0.78, the "within positions or between duplicates" component of variance was 0.07 while the total of the components of variance was 0.85. Thus approximately 92% (91.8%) of the total of the variance components in fat content was due to the positional effect.

It can be seen from Table 20 that the variations in the protein content, both between and within positions were much smaller than in the fat content. Table 23 shows the "Between positions" "F" ratio to be non significant.

These results suggest that the analysis techniques are sufficiently accurate taking into account the variations encountered in the experimental material. It could not be deduced with certainty from these

from these results whether the "positional variations" were due to differences in the fat and protein content of skin taken from different parts of the body, to the inherent variability of the skin fat and protein contents independent of position, or to sampling accidents. For example, the higher value obtained for the fat content in samples F and F₁ could be due to each of the three causes outlined above. Since a correspondingly high value was not obtained for samples C and C₁ taken from the same position on the left side of the sheep, and since in all the skin and wool characters studied a bilateral symmetry exists, it is not likely that this high fat value was due to the presence of gradients in fat content in the area sampled. For example, the accidental inclusion in samples F and F₁ of the subcutaneous fat layer could be responsible for their higher fat content.

TABLE 20

POSITIONAL VARIATIONS AND ERRORS IN THE CHEMICAL
TECHNIQUE

Position	Expressed as % of original weight		
	Fat	Protein	Dry Matter
A	3.3	16.0	19.3
A	3.5	16.3	19.8
B ₁	3.4	16.4	19.8
B ₁	3.5	15.9	19.4
C	3.7	17.9	21.6
C ₁	4.1	15.9	20.0
D	3.0	16.3	19.3
D ₁	3.2	17.2	20.4
E	3.3	17.0	20.3
E ₁	3.8	17.5	21.3
F	5.3	17.0	22.3
F ₁	5.9	17.7	23.6

TABLE 21*

ANALYSIS OF VARIANCE OF THE POSITIONAL VARIATIONS
IN THE FAT CONTENT EXPRESSED AS % OF THE
ORIGINAL WEIGHT AND THE ERRORS INVOLVED IN THE CHEMICAL
TECHNIQUE.

Source of Variations	d.f.	M.S.	F.	Level of Significance
Between positions	5	1.63	23.28	* *
Within positions or between duplicates	6	0.07		
Total	11			

TABLE 22*

ESTIMATES OF THE COMPONENTS OF VARIANCE

Source of variation	d.f.	M.S.	Individuals per subclass	M.S. is an estimate of	Variance Components
Between positions	5	1.63	2	$6_D^2 + 26_P^2$	$6_P^2 = 0.78$
Within positions or between duplicates	6	0.07	1	6_D^2	$6_D^2 = 0.07$

TABLE 23*

ANALYSIS OF VARIANCE OF THE POSITIONAL VARIATIONS
IN THE PROTEIN CONTENT EXPRESSED AS % OF THE ORIGINAL
WEIGHT AND THE ERRORS INVOLVED IN THE CHEMICAL
TECHNIQUE

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between positions	5	0.54	< 1	N.S.
Within positions or between duplicates	6	0.94		
Total	11			

* percentages were analysed as such because they showed a homogeneity of variance.

SUMMARY.

Standard ether extraction and Kjeldahl methods were modified for the determination of fat and protein content of the skin in sheep. Results indicate that:-

1. A period of 24 hours is sufficient for complete fat extraction. A period of 48 hours was henceforth used to ensure complete fat extraction allowing for a wide variation between samples in the rate of extraction.

2. There is no appreciable contamination of fat extracts with protein.

3. Compared with the variation in chemical composition between samples taken from different positions on the sheep, the magnitude of errors due to the methods of determination of fat and protein is not appreciable.

4. Compared with the variation in chemical composition due to the positional variations, the day to day fluctuations in the fat and protein content appear to be small.

C H A P T E R I I

DISTRIBUTION OF SKIN THICKNESS OVER THE BODY OF A SHEEP.

INTRODUCTION.

This experiment was designed to find out how skin thickness varied over the body of a sheep and to identify a uniform sampling area. Such an area is desirable in a study involving more than one sampling per animal in order to prevent the confounding of treatment differences by positional variations in skin thickness. The pelts of three sheep with widely different live weights were sampled in 20 positions and the thickness of the samples was determined.

I. METHODS AND MATERIALS.

(a) Outline of Experiment.

Three sheep of different liveweights were slaughtered. Animals with a wide disparity in size were chosen to discover whether the gradients in skin thickness altered with size. The maximum rectangles permitted by the irregular shapes of the pelts were drawn and each rectangle was further subdivided into 20 (five by four) smaller rectangles. These smaller rectangles are henceforth referred to as "rectangles." A square, one square centimetre in area was marked out in the centre of each rectangle and measured with the Instrument.

Measurements of the pelt dimensions as well as carcase and pelt weights were taken to obtain a more

complete picture of the size of each of the three animals.

(b) Experimental Animals.

One wether hogget (10 months old) in poor condition which had grown badly, one well grown ewe hogget (10 months old), and a three year old wether in store condition, were used in this experiment. The ewe hogget was used because there was no wether hogget of intermediate size available for slaughter. Unfortunately there were no lambs available to compare their skin thickness gradients with those of older animals.

(c) Technique.

The skin thickness measurement technique used was described in Chapter I. The Instrument only was used. Wool was shaved after the samples were removed from the animal and samples were kept in a refrigerator for 24 hours prior to measurement. Each sample was measured five times with the Instrument and the mean obtained.

The rest of the measurements refer to pelts spread horizontally over the floor.

"Shoulder width" S S ' - refers to the measurement taken immediately anterior to the skin covering the front legs.

"Crutch width" K K ' refers to measurement taken immediately anterior to the skin covering the back legs.

"Length" M M ' refers to the measurements taken along the line of the vertebral column from between the ears to the base of the tail.

"Rectangle size" refers to the size of rectangles into which the pelt was divided, the longer side being

parallel to the vertebral column. The figures inscribed in the rectangles, in Diagrams 2, 3 and 4 refer to the means of the skin thickness measurements expressed in millimetres.

II. RESULTS.

The means of the skin thickness measurements are shown in Diagrams 2,3 and 4. The carcase and pelt weights and the pelt dimension measurements appear at the bottom of each of these three Diagrams. Results were presented in this way, no Analyses of Variance being done to test the significance of the differences in skin thickness between the various positions, because the data obtained from three animals of different size and sex were considered insufficient for statistical analysis.

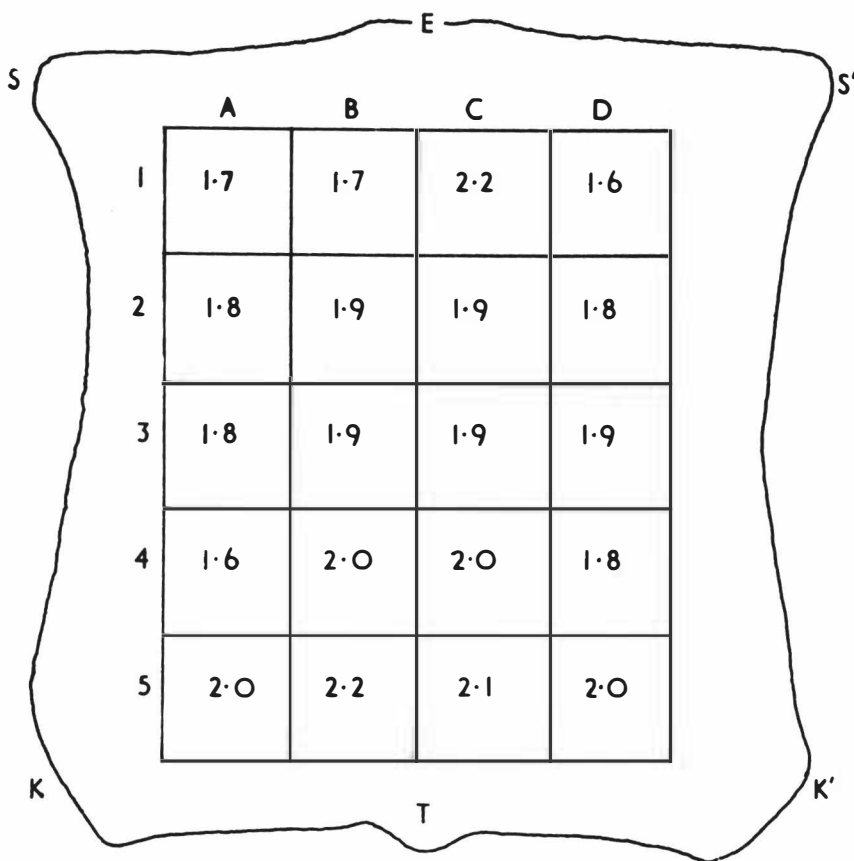
In all three animals, the skin is thicker in the middle of the back (on either side of the vertebral column) than towards the side of the pelt (belly). Likewise, there is a tendency for it to increase in thickness towards the tail. In the older animal (Diagram 4), skin is also thicker in the neck region. There is a uniform area extending on either side of and parallel to the vertebral column, a few inches posterior to the shoulder blades and a few inches anterior to the hip bone (see shaded area in Diagrams 2, 3 and 4).

DISCUSSION.

The general pattern of skin thickness gradients is similar in all three sheep, with the exception of the thicker neck region in the older sheep. Since one ewe and two wethers, a total of three animals only, was used,

DIAGRAM 2

Distribution of skin thickness gradients over the body of a sheep means in mm.

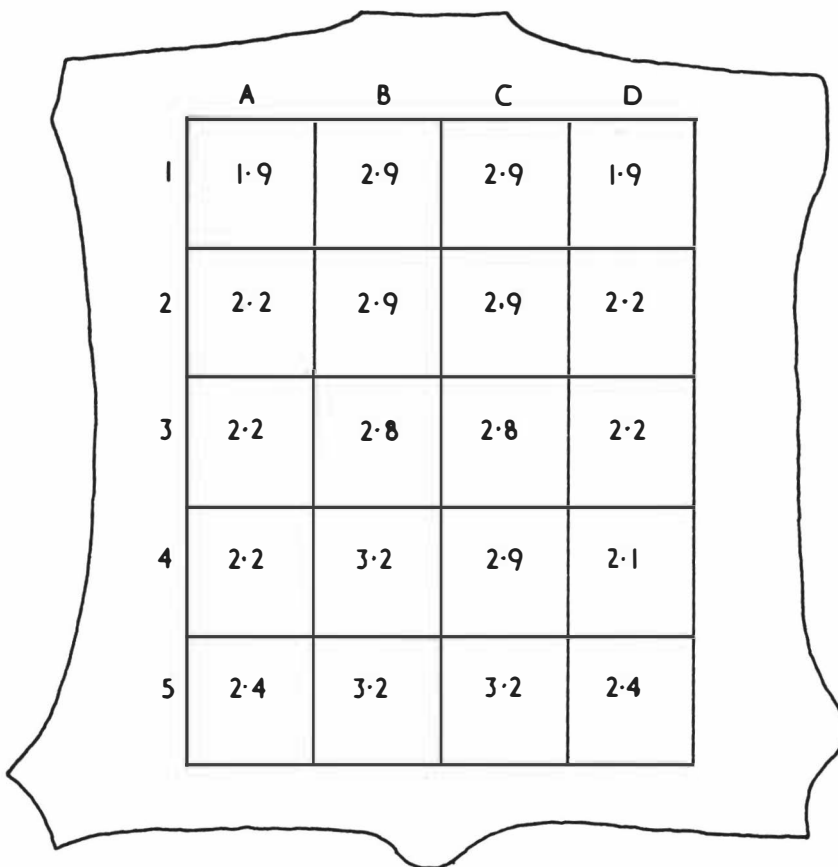


Sheep 1

- Sex Wether
- Age 10 months
- Carcase weight..... 23 lbs.
- Pelt weight..... 5½ lbs.
- Ear-tail (E T)..... 42 ins.
- Shoulder width (SS').. 27 ins.
- Crutch width (KK').. 30 ins.
- Rectangle size..... 5 x 6 ins.

DIAGRAM 3

Distribution of skin thickness gradients over the body of a sheep.
means in mm.



Sheep 2

Sex..... Ewe

Age..... 10 months

Carcase weight..... 53 lbs.

Pelt weight..... 14 lbs.

E T..... 49 ins.

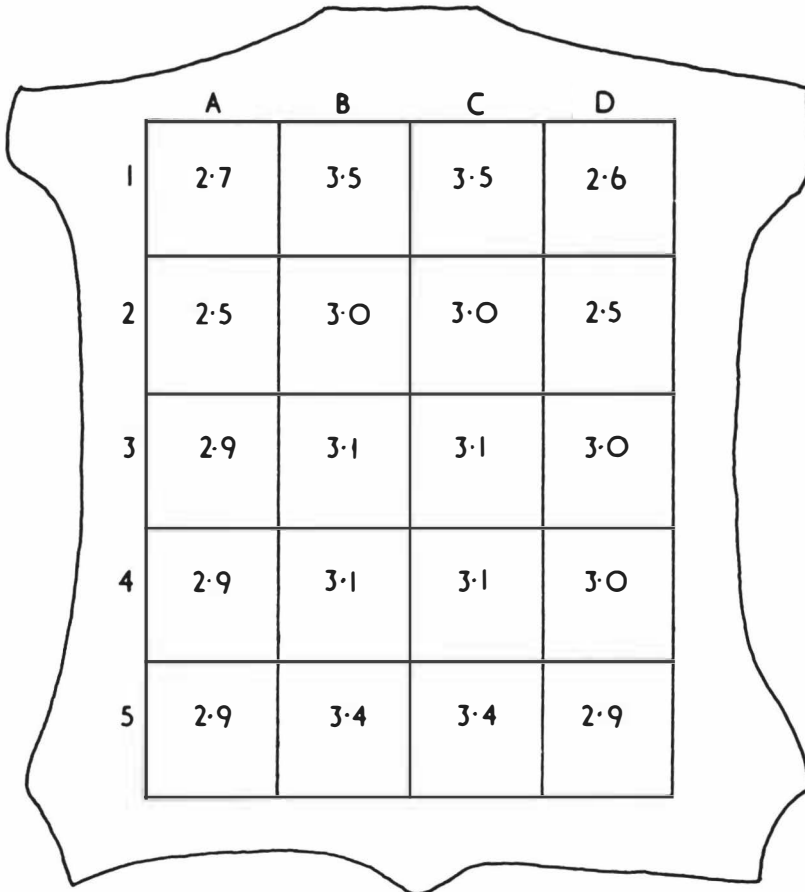
S S'..... 32 ins.

K K'..... 39 ins.

Rectangle size..... 5' 2 x 7 ins.

DIAGRAM 4

Distribution of skin thickness gradients over the body of a sheep means in mm.



Sheep 3

Sex Wether

Age 3 years

Carcase weight 72 lbs.

Pelt weight 15 lbs.

E T 58 ins.

S S' 38 ins.

K K' 48 ins.

Rectangle size 7 x 8 ins.

Galpin (1947) working with the Romney found that the weight of wool produced per square centimetre was higher the nearer the region to the mid-dorsal line. She gives no figures for fibre densities in the different regions, so that it is uncertain whether the greater growth rate of wool in that region is due to the greater fibre density or to the greater mean length grown per day.

If this similarity between gradients in skin thickness, follicle (and may be fibre) density and rate of wool growth is confirmed by future work, it is possible that they are all a reflection of the physiological activity of the skin. Future work on the blood and hence nutrient and hormone supply to the skin (such as that begun by Ryder (1953)), may help to elucidate factors controlling the metabolic activity of the various skin components.

SUMMARY.

Samples were taken from the pelts of three sheep of different live weights to determine the distribution of gradients in skin thickness over the body. Results obtained indicate that:-

1. There is a dorso-ventral gradient in skin thickness, on either side of the median dorsal line which shows a high degree of bilateral symmetry. The skin is thicker near the vertebral column.

2. The skin is thicker near the tail along the back of the sheep, and in the older sheep, also towards the neck.

3. There is an area of uniform skin thickness, in the middle of the back and on either side of and parallel to the vertebral column which is suggested as a suitable sampling area.

C H A P T E R I I I

THE SKIN DEVELOPMENT EXPERIMENT

INTRODUCTION.

Although changes in follicle populations and in skin area during the post-natal period have been studied in the Merino and the Romney by Carter (1943), Carter & Hardy (1947), Burns (1949, 1953), Henderson (1953) and Schinckel (1953), changes in skin thickness and in chemical composition have never been investigated.

In this experiment, changes in the thickness and chemical composition of the skin, occurring from birth to five months, have been studied in 12 Romney lambs.

I. METHODS AND MATERIALS.

(a) Plan of Experiment.

12 Romney wether lambs were sampled within 10 days after birth and at monthly intervals thereafter. At each sampling the skin thickness was measured and the fat and protein contents of the samples determined.

The lambs were weighed and the height at withers measurements were taken at each sampling. Both the liveweight and the height at withers values were obtained because it was considered that the two measurements in combination gave a more complete picture of growth. In addition, the staple lengths

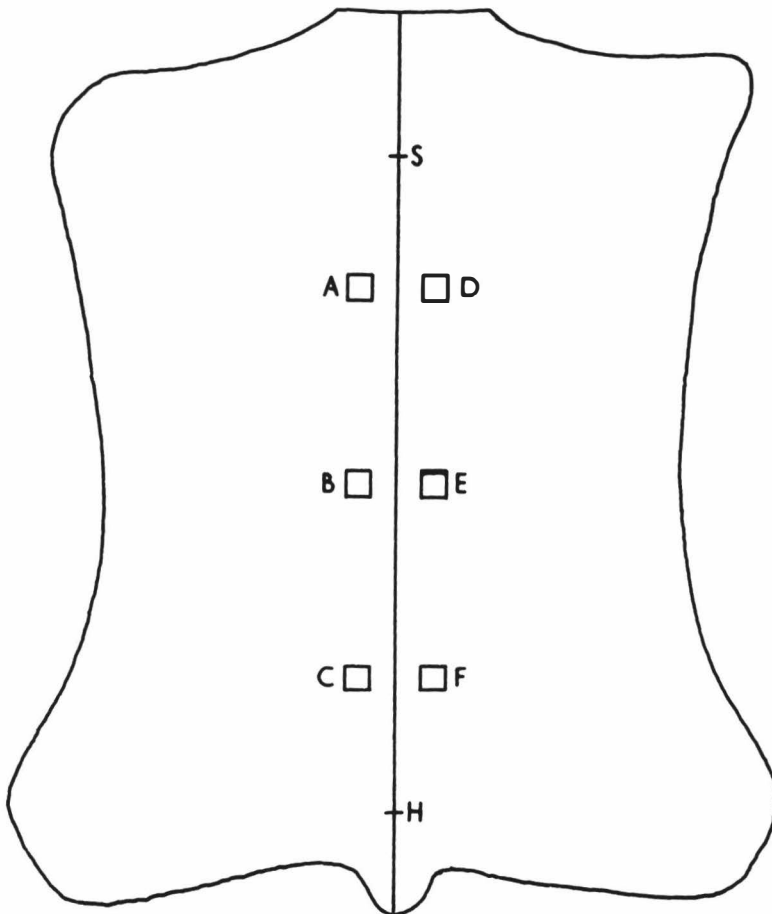
of wool grown post-natally in a position half-way up the side of the animal were measured. No measurement was taken at the first sampling because the latter took place within 10 days after birth when the amount of wool grown post-natally was too small to be measured by the methods employed. At the last sampling, samples of wool were taken from the same position as for the staple length measurement. The wool count or quality was determined and the benzol test for hairiness (McMahon (1937)) carried out. These additional measurements were obtained to see whether they were related to the skin characteristics investigated.

Since it was not known whether the skin thickness, and chemical composition changed rapidly after birth, the first sample should have been taken at a fixed age for all lambs, preferably at birth. Unfortunately, however, the animals were not available until a varying number of days after birth and therefore the first sampling took place one to 10 days after birth.

Results of the experiments described in Chapter II indicated that an area uniform in skin thickness exists parallel to and on either side of the vertebral column, some distance away from the shoulder and hip bones towards the centre of the back. The sampling positions were chosen on the basis of these results and their location on the body of the animal is outlined in Diagram 5. However, this uniform area had been obtained from pelts and was therefore not precisely

DIAGRAM 5

Sampling positions used in the skin development experiment.



S Vertebral column at the shoulder level

H Vertebral column at the hip-bone level

SM = HM

A Sample situated at $\frac{1}{3}$ of the distance SH posterior to S and one inch to the left of the vertebral column.

B Sample situated at $\frac{1}{2}$ of the distance SH posterior to S and one inch to the left of the vertebral column.

C Sample situated at $\frac{1}{3}$ of the distance SH anterior to H and one inch to the left of the vertebral column.

D E & F Samples similarly situated to A B & C but on the right of the vertebral column.

defined on the living animal. Moreover, it was uncertain whether the same gradients in skin thickness occurred in lambs as in older animals.

Thus two variations could confound the time trends in skin thickness and chemical composition - those due to the differences between lambs and those due to the differences between positions within lambs. The Latin square design perfected by Fisher (1925) gives error control in two directions and the two items can be isolated from the error component. For up to eight treatments it is a most efficient type of experiment. As shown in Table 25 two six by six Latin squares were used, the rows denoting the lambs, the columns representing the sampling dates and sampling positions being described by the letters.

(b) Experimental Animals.

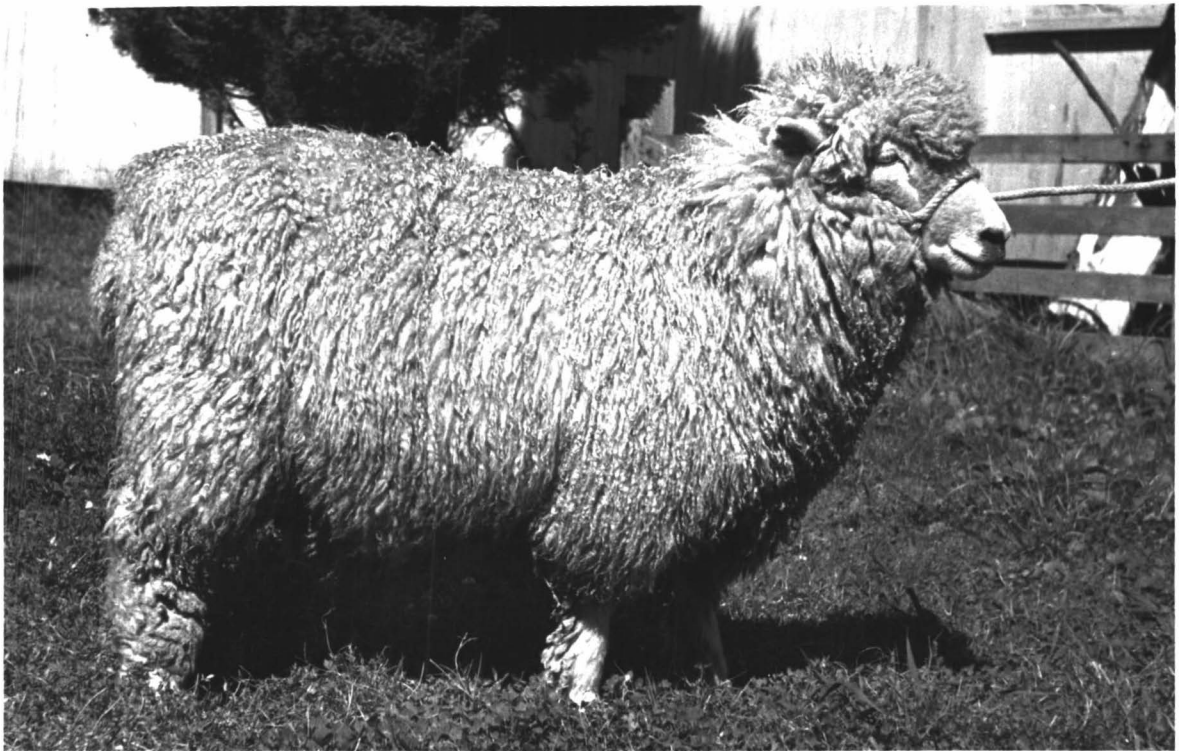
12 single Romney wether lambs were used in this experiment. Photograph 19 shows the lambs at the age of five months. Photograph 20 shows lamb 10 at the age of five months. 10 lambs came from Dr. L.R. Wallace's Fertility Experimental Flock* and two from the Ruakura Hill Country Research Station. The ewes and lambs used in this experiment were run separately from the rest of the Fertility Flock but under similar (i.e. grassland fat lamb) conditions. Like the rest of that flock they were docked at approximately three weeks of age and weaned on 9th December.

Table 24 shows that all lambs except lamb 6 were born between 12th and 30th August. Lamb 6 was a month

* Ruakura Animal Research Station.



Photograph 19: Lambs from the Skin Development Experiment at five months of age.



Photograph 20: Lamb 10 at five months of age.

younger than the rest of the animals, being included in the experimental flock as a result of the death of lamb 6A after the second sampling. Lambs born between the 12th and 18th August (Group A) were sampled on the 20th of each month, lambs born between the 20th and 30th August (Group B) were sampled on the 30th of the month. Lamb 6 was sampled together with Group B, since its first sampling date (within 10 days after birth) came nearer the sampling date of that group.

Lamb 6A died of an overdose of anaesthetic and was replaced, after the second sampling, by lamb 6. Lamb 10 died after the fifth sampling, the suspected cause being pulpy kidney. Lambs 2 and 8 grew at a slower rate than the other animals. Lamb 2 developed, within the first six weeks of its life a very undershot jaw whilst ewe 8 appeared to be a bad milker and dried up completely within two months after lambing. Lamb 5 developed a septic eye following a barb-wire injury shortly after the fourth sampling. It was treated with penicillin, but lost the sight of the left eye and its growth curve showed a check at that point.

Graph 1 and Table 26 show that in spite of being handled twice a month, and subjected once a month to an operation and a separation from their mothers lasting two to four hours, the lambs' growth curves remained normal. Lambs 2 and 8 grew more slowly, but causes other than those due to the experimental procedure are suggested above to be responsible for their slow growth rates. At weaning time these 12 lambs averaged

59 pounds, whilst the Fertility Flock wether lambs averaged 57.5 pounds. Moreover, the proportion of two-tooth (two year old) dams in this experiment was higher than in the Fertility Flock (eight out of 12) and the Fertility Flock two-tooths' wether lambs averaged only 49.5 pounds at weaning time.

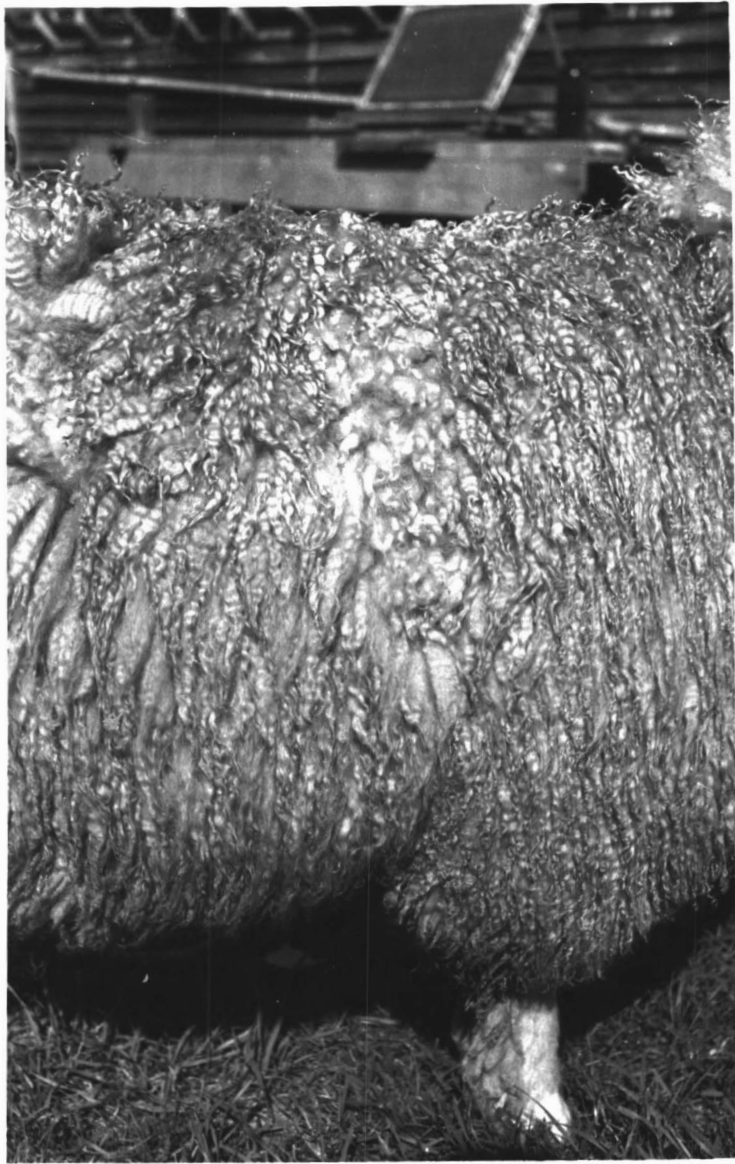
Immediately after the operation the lambs suckled or grazed showing no discomfort. In no case was trouble experienced with the healing of wounds. A month after sampling, the clipped wool and a scar were the only signs of the operation. After three months, the wool had to be completely shaved before the scar marking the site of the operation could be located. Photographs 21, 22, 23 and 24 show the areas sampled at different intervals after the operation.



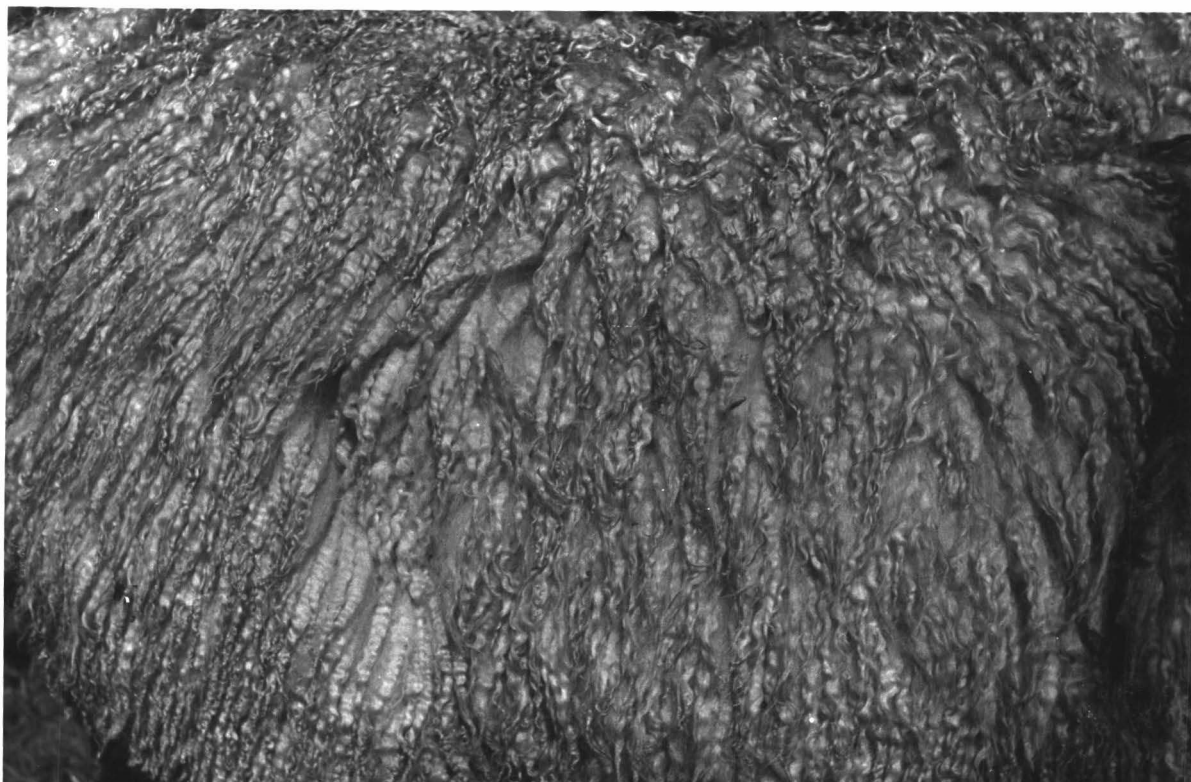
Photograph 21: Area sampled one month after the operation.



Photograph 22: Area sampled two months after the operation.



Photograph 23: Area sampled three months after the operation.



Photograph 24: Area sampled four months after
the operation.

TABLE 24.

EWES AND LAMBS USED IN THE SKIN DEVELOPMENT
EXPERIMENT

Numbers		Age of Ewe	Birth Date of Lamb		Comment
Ewe	Lamb				
1	1	2 yrs.	12th Aug.	1952	Good lamb
2	2	"	13th "	"	Poor lamb with undershot jaw.
3	3	"	16th "	"	Good lamb.
4	4	"	18th "	"	Good lamb.
5	5	"	18th "	"	Good lamb, developed septic eye 20.12.52 & lost some weight.
6*	6*	3 yrs.	24th Sept.	"	Good lamb - replacement for Lamb 6A.
7	7	4 yrs.	20th Aug.	"	Good lamb.
8	8	2 yrs.	23rd "	"	Poor lamb, ewe bad milker.
9*	9*	"	23rd "	"	Good lamb.
10	10	"	23rd "	"	" "
11	11	6 yrs.	27th "	"	" " died 2.1.53, suspected pause pulpy kidney.
12	12	3 yrs.	30th "	"	Good lamb.
6A	6A	2 yrs.	24th "	"	Died 20.9.53.

* from the Ruakura Hill Country Station flock.

TABLE 25

PLAN OF THE SKIN DEVELOPMENT EXPERIMENT.

Sampling dates

Lambs	1	2	3	4	5	6
1	B	F	D	A	E	C
2	D	A	B	C	F	E
3	F	C	E	B	A	D
4	A	E	C	F	D	B
5	E	B	F	D	C	A
6	C	D	A	E	B	F

GROUP A

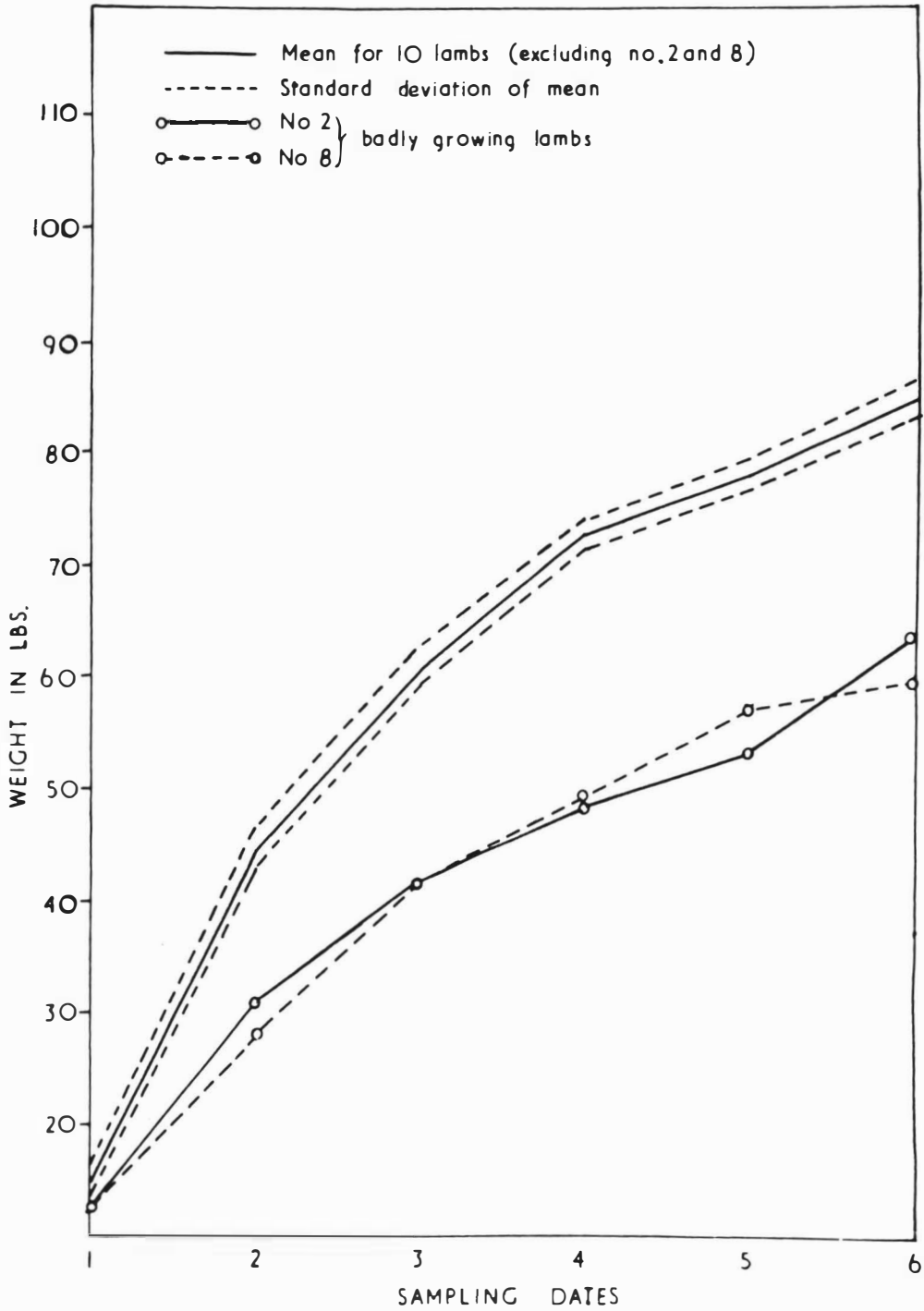
Sampling dates

Lambs	1	2	3	4	5	6
7	D	C	B	F	E	A
8	B	D	E	A	F	C
9	E	F	C	D	A	B
10	A	E	F	B	C	D
11	C	E	A	E	D	F
12	F	A	D	C	B	E

GROUP B

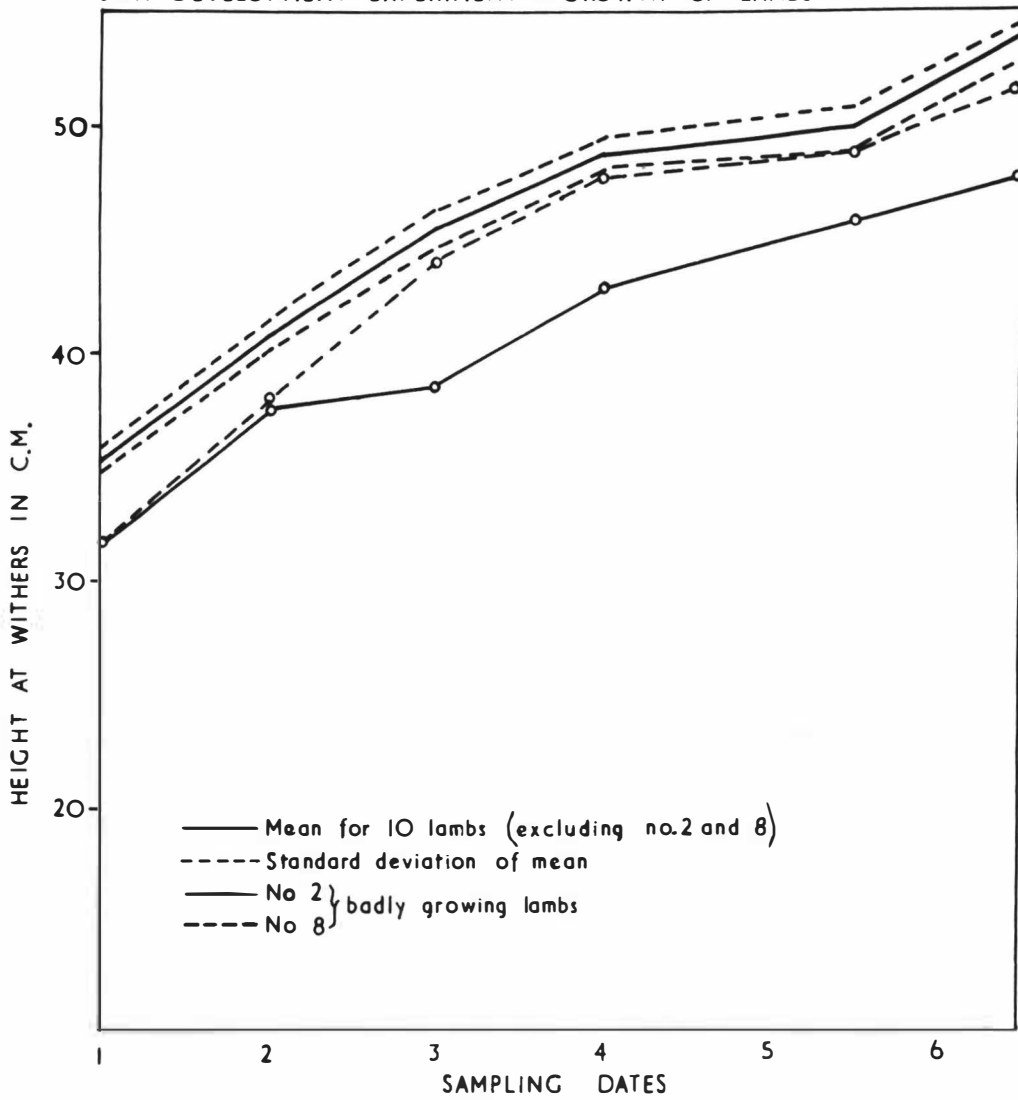
GRAPH 1

SKIN DEVELOPMENT EXPERIMENT GROWTH OF LAMBS



GRAPH 2

SKIN DEVELOPMENT EXPERIMENT — GROWTH OF LAMBS



GRAPH 3

SKIN DEVELOPMENT EXPERIMENT — WOOL GROWTH

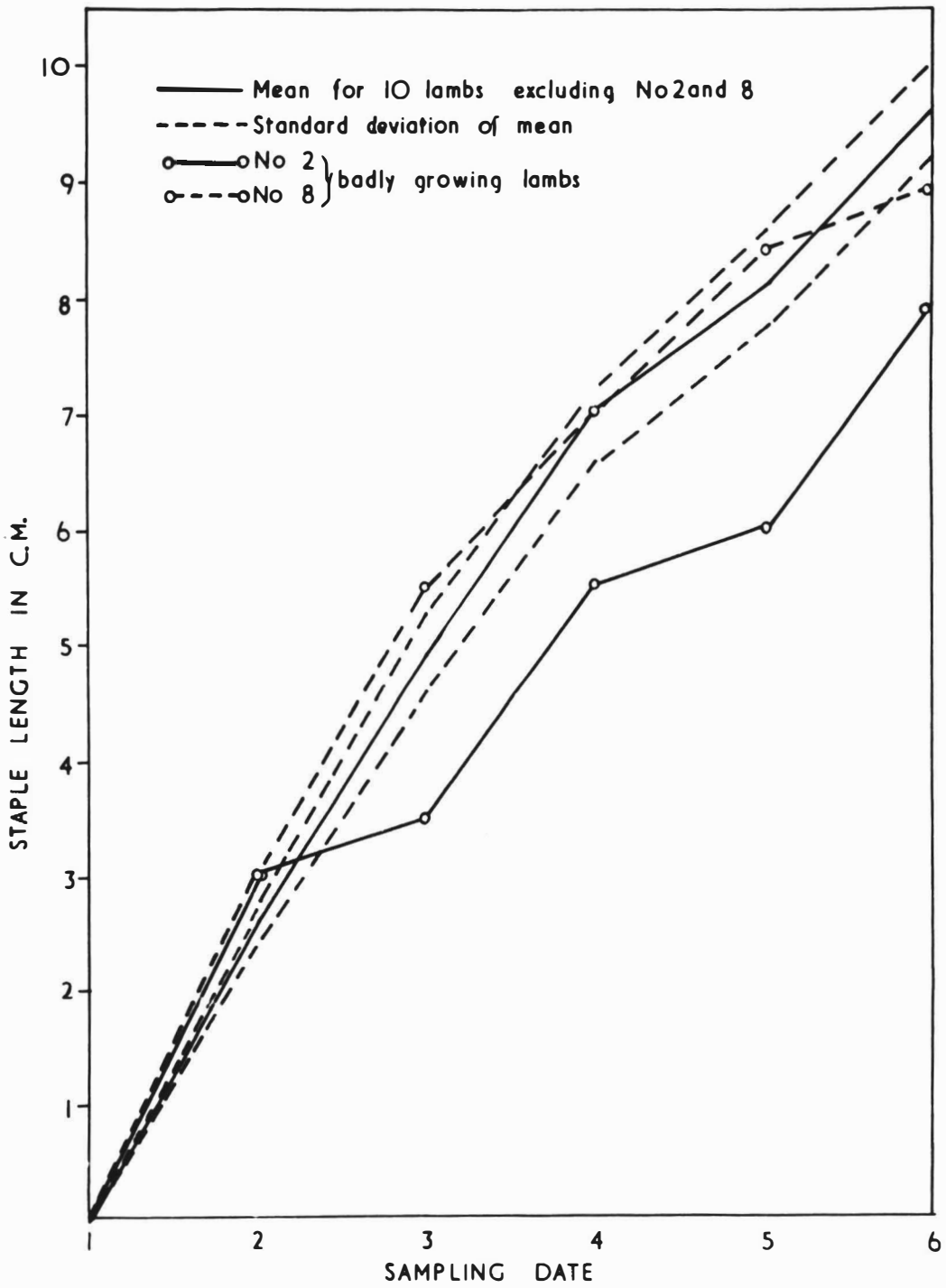


TABLE 26

GROWTH OF LAMBS - WEIGHTS IN POUNDS

	Sampling Dates					
	1	2	3	4	5	6
Mean of 10 lambs excluding lambs 2 and 8	14	34.5	51	63	68	75
Standard deviation	0.9	0.8	0.8	1.0	1.3	1.9
Lamb 2 * *	13	21	32	39	43	54
Lamb 8 * *	13	18	32	39	47	50
Mean of 12 lambs	14	32	47	59	64	71

TABLE 27

GROWTH OF LAMBS - HEIGHT AT WITHERS
(Measurements in pounds)

	Sampling Dates					
	1	2	3	4	5	6
Mean of 10 lambs excluding lambs 2 and 8	36	41	46	49	50	54
Standard deviation	0.5	0.7	0.4	0.4	1.0	0.5
Lamb 2 * *	32	37	38	43	46	48
Lamb 8 * *	36	37	45	48	49	52
Mean of 12 lambs	35	40	45	48	50	53

* * badly growing lambs.

(c) Experimental Technique.

The technique adopted for the determination of skin thickness and chemical composition was similar to the one described in Chapter I (A) and (B). However, the local anaesthetic was not used prior to the operation, since it was feared that it might affect the thickness and chemical composition of the skin either through the direct addition of extraneous matter or by its differential effect on the various skin components. Total anaesthesia produced by sodium pentathol given intravenously (into the jugular vein) at the rate of one gram per seven pounds live-weight, was used at the first two samplings. However, after lamb 6A died of an overdose of the anaesthetic, its use was discontinued.

The skin was shaved before being removed from the animal, since it was feared that excessive handling after removal might alter its thickness. Although experiments described in Chapter I (A) showed that in older animals skin could be shaved after removal from the sheep without altering in thickness, it was thought that the skin of lambs might be thinner and less resistant to handling. Samples were kept for 24 hours in a refrigerator prior to being measured with the Instrument. Each sample consisted of four equal squares, each square being one square centimetre in area. Each square was measured five times and the mean of the 20 measurements per sample was calculated. The chemical analysis procedure was identical with the one described in Sections I (a) and (b) of Chapter I (B).

The staple lengths of wool were measured in a position halfway up the side of the animal, from the skin level to the base of the "curly tip" (i.e. the wool grown pre-natally). The staple was laid along a ruler without being stretched and its length was measured to the nearest half centimetre. Three staples were measured and the mean was obtained. The mean was found to vary by no more than ± 0.5 centimetre within one animal on a given sampling date. The wool count or quality and the percentage hairiness were determined on a wool sample removed from the same position on the animal as for the staple length measurement. The count was determined by the eye assessment method used in commercial wool classing. The percentage hairiness was determined by the benzol test developed by McMahon (1937) in which the discrimination between medullated (or hairy) and non medullated fibres depends on the different refractive index of the two types of fibres.

The lambs were weighed, to the nearest pound at each sampling, on a spring balance during the first two times and on Avery automatic scales the other four times. The height at withers measurement was taken against a wall, with a pair of measuring calipers, an accuracy of + one centimetre being attained.

(d) Sampling Procedure.

The lambs were separated from the ewes, weighed, measured and sampled. Two operators were needed. During the last four samplings when no anaesthetic was used, the lamb was tied and held by one operator to

prevent it from struggling. The first sampling took place in the paddock to make the separation of the lambs from the ewes as short as possible. In subsequent samplings the lambs were taken to the woolshed where instruments could be sterilized and sampling was more convenient. The weighing, and the wool and body measurements took about 30 minutes per group (of six lambs), the remainder of the operation about 10 minutes per lamb, although the actual skin removal took only about 30 seconds.

II. RESULTS.

(a) Skin Thickness.

Changes in skin thickness during the experimental period are shown in Table 28 and Graph 4. Mean skin thickness values obtained at each sampling are given for all 12 lambs and for 10 lambs, excluding lambs 2 and 8 which grew more slowly than the remaining animals. Skin thickness values for lambs 2 and 8 are given separately.

Table 28 and Graph 4 show that the skin increased in thickness from the first sampling (one to 10 days after birth) to the second sampling (30 to 40 days after birth). After the third sampling (eight to 10 weeks after birth) skin thickness decreased to a value similar to the one obtained at the first sampling. From the fourth sampling (12 to 14 weeks after birth) until the end of the experiment when the animals were approximately five months of age, skin thickness remain unchanges. Table 29 shows that the "F" ratio for "sampling dates" was significant at the 1% level.

GRAPH 4

SKIN DEVELOPMENT EXPERIMENT — SKIN THICKNESS

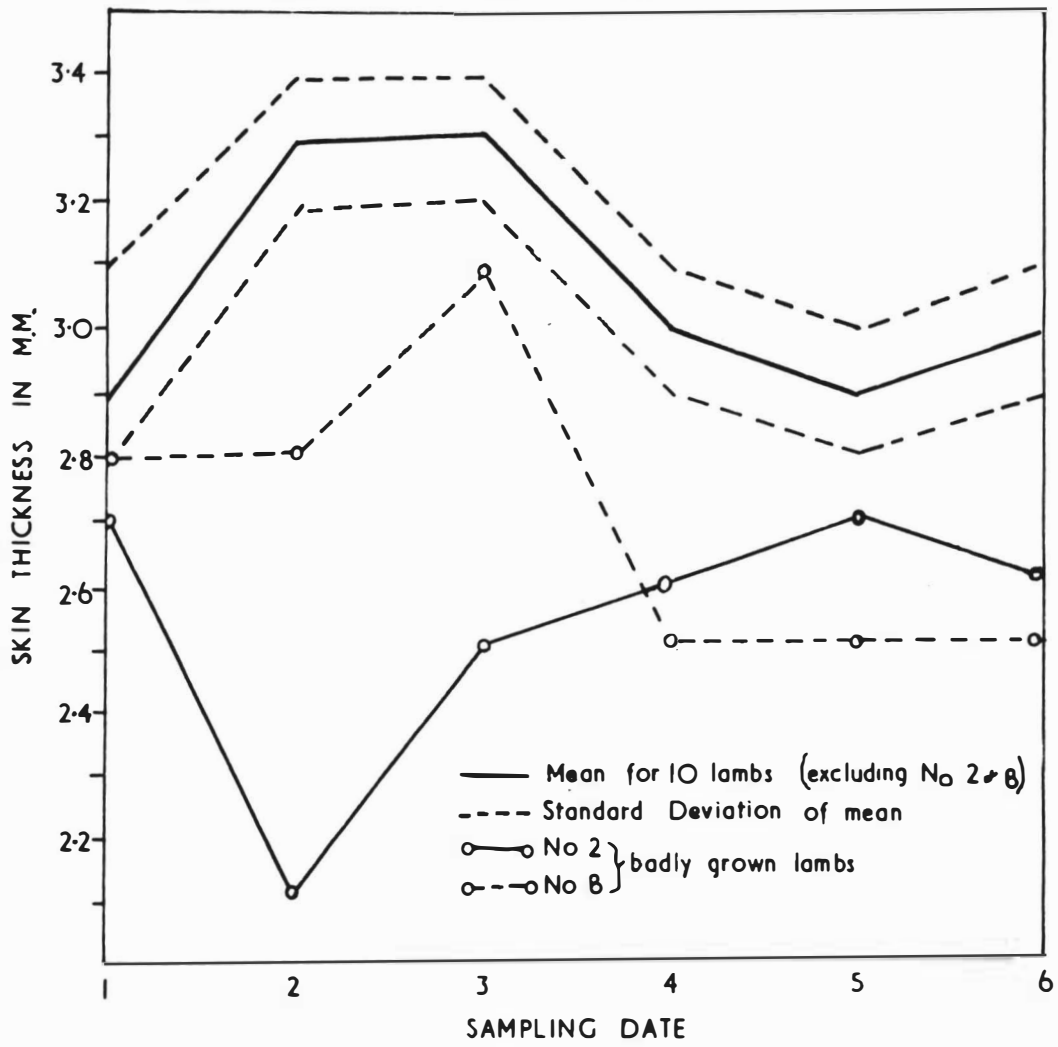


TABLE 28.

CHANGES IN SKIN THICKNESS
(Millimetres)

	SAMPLING DATES					
	1	2	3	4	5	6
Mean of 10 lambs excluding lambs 2 and 8	2.9	3.3	3.3	3.0	2.9	3.0
Standard deviation	0.2	0.1	0.1	0.1	0.1	0.1
Lambs 2 * *	2.7	2.1	2.5	2.6	2.7	2.6
Lambs 8 * *	2.8	2.9	3.1	2.5	2.5	2.5
Mean of 12 lambs	2.8	3.2	3.2	2.9	2.8	2.8

TABLE 29.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED
IN TABLE 28.

(For convenience, all calculations for the Analysis of Variance in this Table are in tenths of a millimetre)

SOURCE OF VARIATION	d.f.	M.S.	F.	Level of significance
Between Latin squares	1	260.68	< 1	N.S.
" lambs within Latin squares	10	15,698.75	6.24	* *
Between positions	5	15,682.98	6.24	* *
left v. right side of the body	1	550.01	< 1	N.S.
linear regression along the length of the body	1	41,713.02	16.59	* *
quadratic regression along the length of the body	1	154.17	< 1	N.S.
side x length	2	17,998.85	7.16	* *
Positions x Latin squares	5	510.36	< 1	N.S.
Between sampling dates	5	12,502.01	4.97	* *
Sampling dates x Latin squares	5	7,563.08	3.01	*
Error	39	2,514.49		
Total	70			

Table 28 and Graph 4 show that lambs 2 and 8 which grew more slowly than the other 10 animals, had thinner skins. Their skin thickness values after the first sampling, fell below the mean and lay outside the standard deviation limits for the remainder of the experimental group. Lamb 8 had a skin thickness curve similar to the mean curve for the other 10 animals, though the values at each sampling were lower. The curve of lamb 2, besides having lower values throughout, did not show a peak at the two to three months period.

There was no correlation between skin thickness and age, liveweight, liveweight gain, or height at withers over all sampling dates, because while skin initially increased in thickness, then decreased and finally remained at the same value until the end of the experiment, the other measurements continued to increase throughout. In Table 30, correlations at each sampling date (within sampling dates) between skin thickness and age, and skin thickness and growth as measured by liveweight, liveweight gain, relative growth rate* and height at withers, are given.

There was a correlation between skin thickness and liveweight (both corrected and uncorrected for age) which was significant at the 1% level, at the first,

$$* \text{ relative growth rate} = K = \frac{dw/dt}{w}$$

where dw/dt = gain in liveweight per unit time (one month)

and w = weight of the animal at the instant the rate dw/dt is measured. (Brody 1945)

fourth and sixth samplings, and significant at the 5% level at the second sampling. A method given by Snedecor (1946) was used to test the hypothesis that the correlations between skin thickness and liveweight within sampling dates came from the same population and could be combined into an estimate of an overall correlation. Each r was converted into Z (Fisher & Yates (1953) Table V) and each Z was weighted by the reciprocal of its variance. From the sum of the weighted Z 's, a correction factor was subtracted and the resultant chi-square tested. The chi-square value fell between $p = 0.30$ and $p = 0.50$. It was therefore concluded that there was no reason to suspect that the six correlations might not have been drawn from the same population. Their average value obtained by transforming the average Z to r was found to have a value of 0.707 which was significant at the 1% level.

There was a correlation, significant at the 1% level between skin thickness and liveweight gain at the first, third, fourth and sixth samplings, whilst the correlation approached significance at the 5% level at the second and sixth samplings. Using the methods outlined in the last paragraph, it was shown that these correlations came from the same population and that the estimated overall correlation had a value of 0.799, which was significant at the 1% level.

There was, however, no significant correlation, between skin thickness and relative growth rate.

Except for the second sampling, no significant correlation existed between skin thickness and the height at withers.

At the first sampling there was a correlation, significant at the 1% level between skin thickness and age. At the other five samplings, however, no significant correlation existed between skin thickness and age within samplings. Since there was a correlation, significant at the 1% level between weight and age at the first sampling, it is suggested that liveweight and liveweight gain differences rather than age differences within the experimental group were responsible for the differences in skin thickness between lambs at a given sampling date.

Although it is dangerous to generalise on the basis of results obtained from 12 animals, it seems that skin thickness from birth to five months follows a characteristic curve, but that within this curve the bigger, more rapidly growing lambs have thicker skins.

Sampling Date	d.f.	Weight-Age	Level of Significance	Thickness-Age	Level of Significance	Thickness-Live-weight	Level of Significance	Weight Thickness-Age (partial correlation coefficient)	Level of Significance	Thickness-Live-weight gain	Level of Significance	Thickness-Relative Growth Rate	Level of Significance	Thickness-Height at withers	Level of Significance
1	10	0.808	* *	0.668	* *	0.929	* *	0.888	* *	0.818	* *	0.219	N.S.	-0.155	N.S.
2	10	0.130	N.S.	0.374	N.S.	0.678	*	0.684	*	0.524	N.S.	0.248	N.S.	0.683	* *
3	10	0.027	N.S.	0.380	N.S.	0.508	N.S.	0.560	N.S.	0.723	* *	0.339	N.S.	0.414	N.S.
4	10	0.192	N.S.	0.360	N.S.	0.780	* *	0.777	* *	0.818	* *	0.569	N.S.	0.550	N.S.
5	10	0.320	N.S.	0.109	N.S.	0.446	N.S.	0.511	N.S.	0.967	* *	0.194	N.S.	0.085	N.S.
6	9	0.171	N.S.	0.254	N.S.	0.778	* *	0.771	* *	0.551	N.S.	0.360	N.S.	0.387	N.S.

TABLE 30.

CORRELATIONS BETWEEN SKIN THICKNESS AND GROWTH WITHIN SAMPLING DATES

The growth of wool as represented by the staple length measurements in centimetres is shown in Table 31 and Graph 3. Mean values obtained at each sampling are given for all 12 lambs and also for the 10 lambs excluding lambs 2 and 8. There was no correlation between skin thickness and staple length between sampling dates because while skin initially increased in thickness, then decreased and finally remained at the same value until the end of the experiment, the other measurements continued to increase throughout. As shown in Table 32, there was no significant correlation between skin thickness and staple length within sampling dates.

TABLE 31.

GROWTH OF WOOL - STAPLE LENGTH IN
CENTIMETRES.

		<u>Sampling Date</u>				
		2	3	4	5	6
Mean	of 10 lambs excluding lambs 2 and 8	2.6	4.9	6.9	8.2	9.7
Standard deviation	of lambs 2 and 8	0.1	0.3	0.3	0.4	0.4
Lamb 2	**	2.0	3.5	5.5	6.0	8.0
Lamb 8	**	3.0	5.5	7.0	8.5	9.0
Mean	of 12 lambs	2.6	4.8	6.8	8.1	9.5

** badly growing lambs

TABLE 32.

CORRELATIONS BETWEEN SKIN THICKNESS AND
STAPLE LENGTH WITHIN SAMPLING DATES

Sampling Date	d.f.	r Thickness· Staple Length	Level of Signifi- cance
2	10	0.397	N.S.
3	10	-0.090	N.S.
4	10	0.255	N.S.
5	10	0.029	N.S.
6	9	0.046	N.S.

In Table 33 the mean values for the percentage hairiness and the count or quality of wool obtained at the sixth sampling are given for each of the 12 lambs together with the skin thickness measurements obtained at the sixth sampling and the means of all the skin thickness measurements obtained throughout the experiment.

As shown in Table 34 this experiment did not reveal significant correlations between skin thickness and percentage hairiness of wool and skin thickness and wool count.

TABLE 33.

% HAIRINESS COUNT OR QUALITY AND
SKIN THICKNESS.

Lamb Number	% Hairiness	Count or Quality	Skin Thickness (millimetres)	
			Sixth Sampling	Mean
1	4.20	50	3.4	3.3
2**	2.00	56	2.6	2.5
3	2.55	52	3.0	3.0
4	1.65	50/48	2.9	3.0
5	4.65	54	2.9	3.0
6	2.10	48/50	3.0	3.1
7	7.25	52	2.8	3.0
8**	1.85	50	2.5	2.7
9	3.00	48	2.8	2.9
10	1.60	54	3.0	3.1
11*				
12	4.60	50/48	2.5	2.7
Mean	3.20	50	2.8	2.9

* died before the sixth sampling.

** badly growing lambs.

TABLE 34.

CORRELATIONS BETWEEN SKIN THICKNESS AND % HAIRINESS
AND SKIN THICKNESS AND COUNT OR QUALITY.

Thickness, hairiness and count at the sixth sampling.	d.f.	Level of Significance	Mean thickness; hairiness and count at the sixth sampling	d.f.	Level of Significance
r Thickness·hairiness	0.029	9	N.S.	r Thickness·hairiness	0.163 9 N.S.
r Thickness·count	-0.044	9	N.S.	r Thickness·count	-0.238 9 N.S.

Mean values for the thickness and fat, protein and dry matter contents of skin at each of the six positions are given in Table 35.

Table 29 shows that the differences between positions were significant at the 1% level. The separation of the mean square for "positions" into its components showed that the "F" ratios for the "linear regression along the length of the body" and the "side x length" interaction were significant at the 1% level. As seen in Table 35, on the left side of the sheep there was a postero-anterior gradient in skin thickness, but on the right side, although the thickest skin was found in the back position, the middle position had lower values for skin thickness than the front position.

Since all characteristics of the skin of sheep so far studied, show a high degree of bilateral symmetry, it would seem that these differences between positions were not due to the inherent differences in skin thickness at the corresponding positions, but rather to errors in the sampling and/or measurement techniques.

Results from the Preliminary Experiment I (Chapter IA) indicated that the same number of cell layers were removed at each sampling and the fact that in this experiment the positional differences in skin thickness did not correspond to those in the fat content, suggest that inclusion of varying amounts of subcutaneous fat was not the cause of increased skin thickness at a given sampling position. Repeat measurements showed that the Instrument measurements were highly accurate. It

is suggested therefore that these differences between positions were due rather to the inaccurate defining of the sampling positions. As results from the experiment described in Chapter II indicated that a uniform sampling area existed some distance away from the shoulder and hip bones, parallel to and an inch or two on either side of the vertebral column, these were the sampling positions used for the Skin Development Experiment. However, results of the experiments described in Chapter II showed that the main gradient in skin thickness was dorso-ventral and therefore a small variation between samples in the distance from the vertebral column would lead to a big difference in the skin thickness values obtained. It is likely that, because it is more difficult to judge exactly distances from the vertebral column on a live animal than on a pelt, accidental variations between samples in the distance from the vertebral column were largely responsible for the between positions variation in skin thickness.

Although the Latin square design prevented confounding of the changes in skin thickness with increasing age by the positional differences, it is suggested that in future work, samples should be taken closer to the middle of the back, to avoid the possible postero-anterior gradient in skin thickness, and immediately adjacent to the vertebral column, to avoid errors due to the presence of a steep dorso-ventral gradient.

TABLE 35.

POSITIONAL DIFFERENCES IN THE THICKNESS, FAT
CONTENT AND DRY MATTER CONTENT OF SKIN - MEANS OF
12 LAMBS IN MILLIMETRES.

Position	Left	Right
<u>Front</u>		
Thickness (millimetres)	2.75	3.0
Fat content (expressed as % of the original material)	2.7	3.5
Protein content (ditto)	13.6	14.8
Dry matter content (ditto)	16.7	18.7
<u>Middle</u>		
Thickness (millimetres)	3.15	2.9
Fat content (expressed as % of the original material)	3.9	3.1
Protein content (ditto)	14.0	14.3
Dry matter content (ditto)	18.3	17.8
<u>Back</u>		
Thickness (millimetres)	3.3	3.1
Fat content (expressed as % of the original material)	3.5	3.2
Protein content (ditto)	14.7	14.4
Dry matter content (ditto)	18.5	17.9

(b) Chemical.

Changes, during the experimental period, in the fat protein and dry matter contents of the skin, expressed as percentages of the original weight are shown in Tables 36, 40 and 42 and Graphs 5, 7 and 8. Changes in the fat content expressed as percentage of the total dry matter are shown in Table 38 and Graph 6. Mean values obtained at each sampling are given for all 12 lambs and for the 10 lambs, excluding lambs 2 and 8. Chemical composition values for lambs 2 and 8 are given separately in each Table.

The protein and dry matter percentages were analysed as such because they did not appear to violate the conditions required for the Analysis of Variance. These conditions are:- firstly, normality at the residual error term; secondly, homogeneity of the residual error term; and thirdly, absence of correlation between the residual error term. Bartlett's test* in particular showed that the variances were homogeneous.

The fat content of skin expressed both as percentage of the original material and as percentage of the total dry matter showed a variance which increased from the first to the last samplings. Bartlett's test for the homogeneity of variance was applied and the corrected chi-square was found to be 23.93 for the fat content expressed as percentage of the original material and 14.10 for the fat content expressed as percentage of the total dry matter, showing both variances to be non homogeneous, the former at the 1% and the latter at the 5% level of significance. The arc sin and the logarithmic transformations were tried on the data. The logarithmic transformations were found to be the most satisfactory in reducing the ratio of the biggest to the smallest variance from 26.29 to 5.38 in the fat percentage of original weight series, and from 9.85 to 1.33 in the fat percentage of dry matter series. All percentages therefore, for the fat content calculations were transformed to logarithms before analysis.

In the untransformed data presented in this chapter (skin thickness, protein and dry matter percentage) the means and standard deviations at each sampling

* Snedecor (1946)

date are given. In the fat content data, the means and confidence limits are given because the antilogarithms obtained for the upper and lower limits are found to be situated at an unequal distance from the mean.

Tables 36 and 38 and Graphs 5 and 6 indicate that the fat content of skin expressed either as percentage of the original weight or as percentage of the total dry matter rose with increasing age. Table 37 and 39 show these "F" ratios for "sampling dates" to be significant at the 1% level.

Tables 36 and 38 and Graphs 5 and 6 indicate that lambs 2 and 8 which grew more slowly than the other 10 animals, had skins with a lower fat content. The fat contents, except for the fifth sample from lamb 2 (fat content expressed as percentage of the original weight) and the third and fifth sample from lamb 2 (fat content expressed as percentage of the total dry matter), fell below the means for the rest of the experimental group. The fat contents of lambs 2 and 8 did not, however, lie outside the 95% confidence intervals. There was a correlation of 0.653 (significant at the 1% level) between fat content and liveweight over all the sampling dates.

GRAPH 5

SKIN DEVELOPMENT EXPERIMENT

FAT CONTENT EXPRESSED AS % OF THE ORIGINAL WEIGHT.

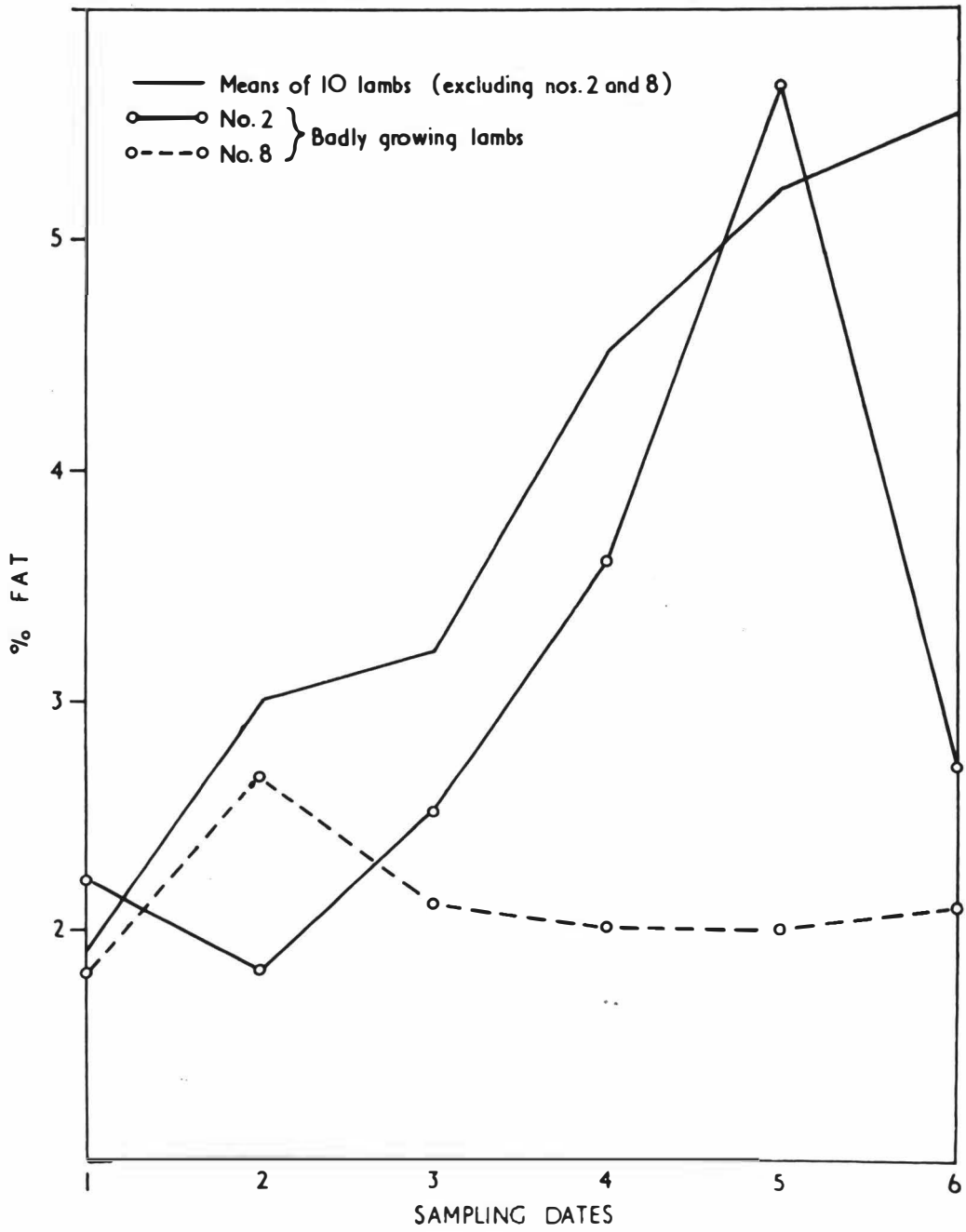


TABLE 36

CHANGES IN THE FAT CONTENT OF SKIN EXPRESSED
AS % OF THE ORIGINAL WEIGHT.

		<u>Sampling Date</u>					
		1	2	3	4	5	6
Mean	of 10 lambs	1.9	2.9	3.1	4.3	4.8	5.2
95% Confidence Intervals	lower excluding upper lambs 2 & 8	0.6	0.6	0.6	0.5	0.4	0.7
		6.3	13.4	16.05	36.0	59.9	40.1
Lamb 2**		2.2	1.8	2.5	3.6	5.4	2.7
Lamb 8**		1.8	2.7	2.1	2.0	2.0	2.1
Mean of 12 lambs		1.9	2.8	2.9	3.95	4.5	4.6

* * badly growing lambs.

TABLE 37.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED IN

TABLE 36***

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin Squares	1	0.0030	< 1	N.S.
Between lambs within Latin squares	10	0.1059	21.61	* *
Between positions within Latin squares	10	0.0254	5.18	* *
Between dates	5	0.2604	53.14	* *
Dates x Latin squares	5	0.0129	2.63	*
Error	39	0.0049		
Total	70			

*** Percentages transformed into logarithms for all calculations in this Table.

GRAPH 6

SKIN DEVELOPMENT EXPERIMENT

FAT CONTENT EXPRESSED AS % OF THE TOTAL DRY MATTER

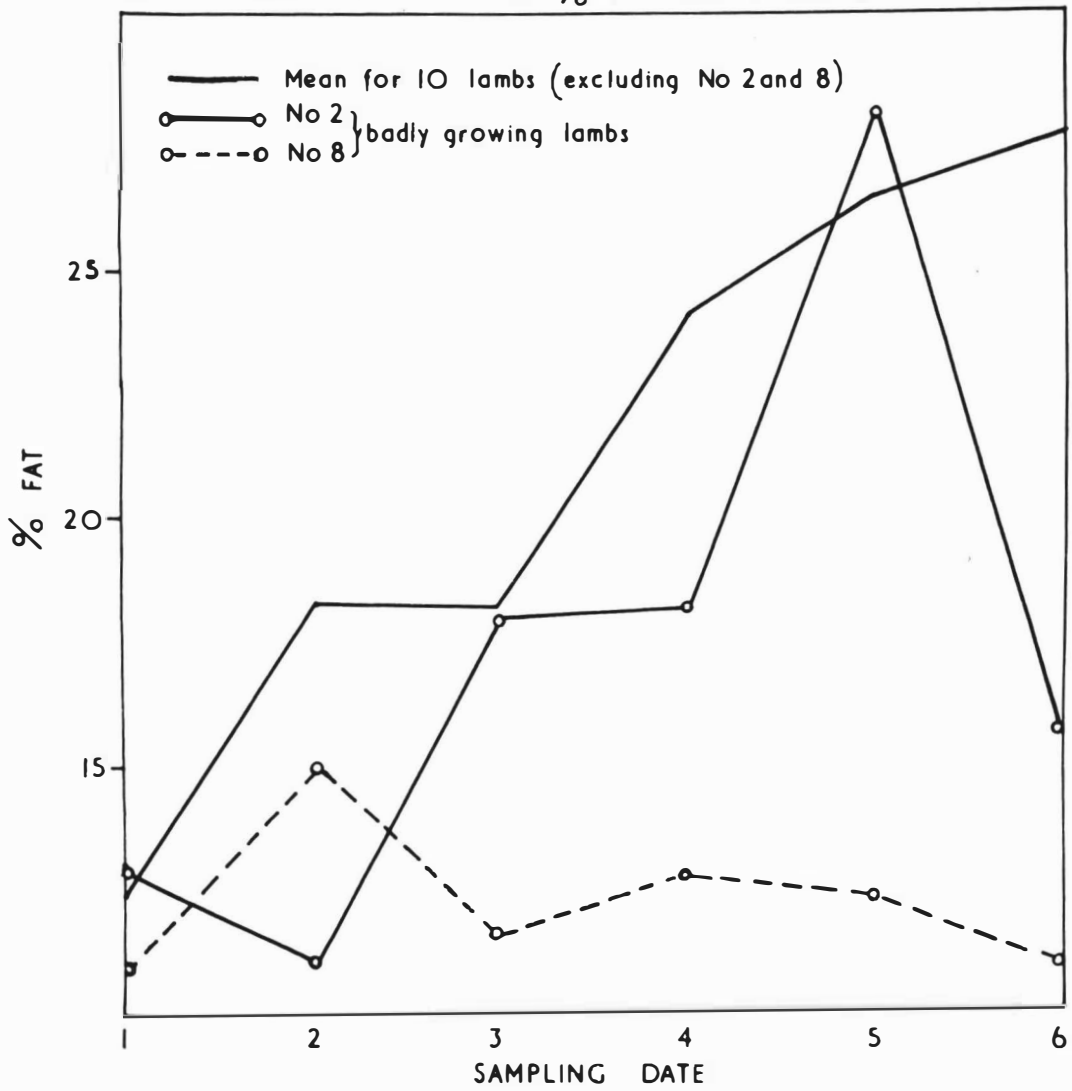


TABLE 38.

CHANGES IN THE FAT CONTENT OF SKIN EXPRESSED
AS % OF THE DRY MATTER.

			<u>Sampling Date</u>					
			1	2	3	4	5	6
Mean		of 10 lambs	12.0	17.5	17.5	23.0	15.2	27.5
95% Confidence Intervals	lower	excluding lambs 2 and 8	3.6	4.4	4.2	3.3	2.9	5.4
	upper		39.6	69.2	71.8	161.7	220.7	139.8
Lamb 2 **			12.8	11.0	17.8	18.0	28.2	15.4
Lamb 8 **			10.5	14.8	11.5	12.8	12.4	10.9
Mean of 12 lambs			11.9	16.6	16.9	21.4	24.0	24.2

TABLE 39.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED
IN TABLE 38 ***

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	1	0.0048	< 1	N.S.
Between lambs within Latin squares	10	0.0699	12.71	* *
Between positions within Latin squares	10	0.0248	4.51	* *
Between dates	5	0.1693	30.78	* *
Dates x Latin squares	5	0.0126	2.29	N.S.
Error	39	0.0055		
Total	70			

*** Percentages transformed into logarithms for all calculations in this Table.

GRAPH 7

SKIN DEVELOPMENT EXPERIMENT

PROTEIN CONTENT EXPRESSED AS % OF ORIGINAL WEIGHT

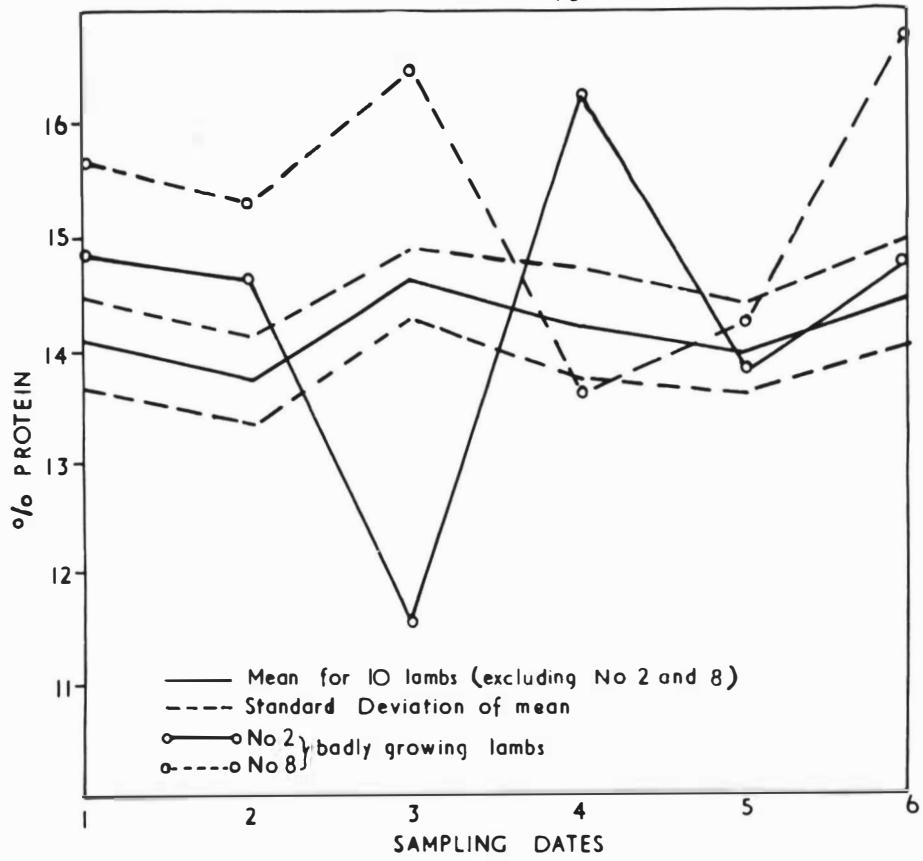


Table 40 and Graph 7 show that the protein content of the skin did not alter appreciably over the experimental period. Table 41 shows that the "F" ratio for "sampling dates" was non significant. As shown in Table 40 and Graph 7, the mean protein content, over the experimental period, of the skin from the two badly growing lambs did not differ from that of the other 10 animals.

TABLE 40.

THE PROTEIN CONTENT OF SKIN EXPRESSED AS %
OF THE ORIGINAL WEIGHT.

		<u>Sampling Dates.</u>						
		1	2	3	4	5	6	Mean
Mean	of 10 lambs excluding lambs 2 and 8	14.1	13.8	14.6	14.2	14.0	14.5	14.2
Standard deviation		0.4	0.4	0.3	0.55	0.45	0.5	
Lamb 2**		14.8	14.7	11.5	16.3	13.8	14.8	14.3
Lamb 8**		15.7	15.4	16.5	13.6	14.2	16.9	15.4
Mean of 12 lambs		14.2	14.0	14.5	14.3	14.0	14.9	14.3

** badly growing lambs

TABLE 41.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED IN TABLE 40.

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	1	2.03	1.40	N.S.
Between lambs within Latin squares	10	5.16	3.56	* *
Between positions within Latin squares	10	1.77	1.22	N.S.
Between dates within Latin squares	10	2.02	1.39	N.S.
Error	39	1.45		
Total	70			

GRAPH 8

SKIN DEVELOPMENT EXPERIMENT

DRY MATTER CONTENT EXPRESSED AS % OF THE ORIGINAL WEIGHT.

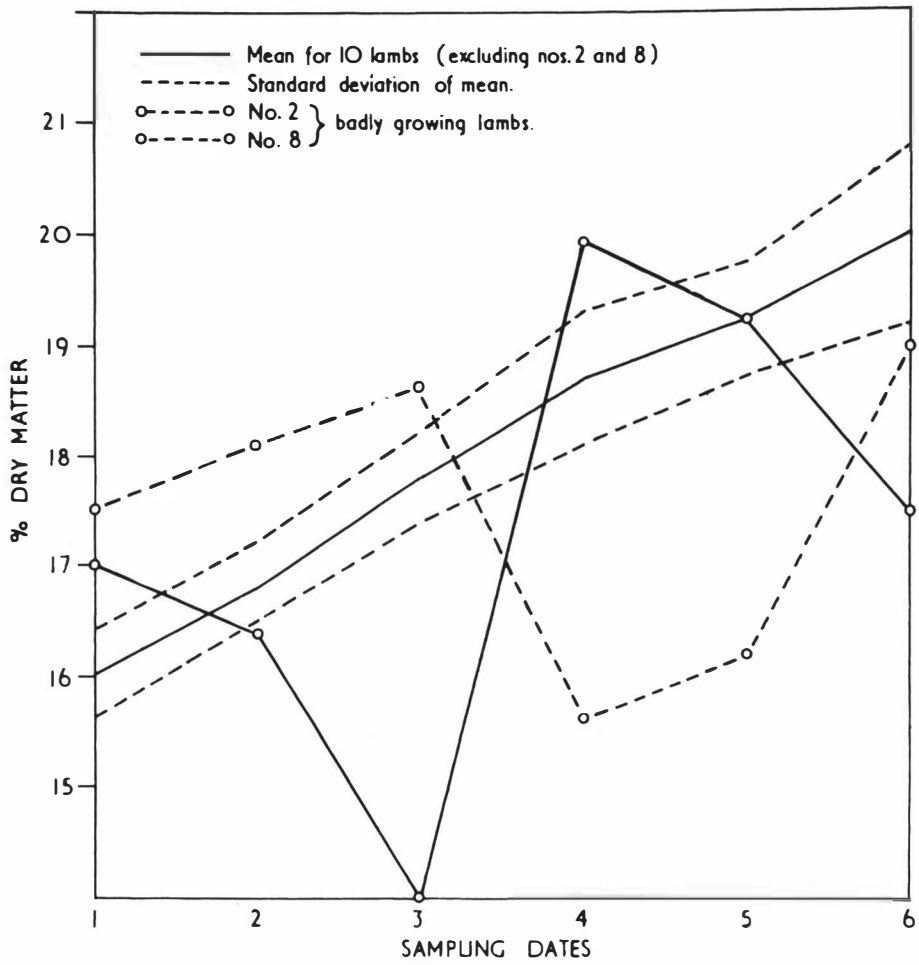


Table 42 and Graph 8 indicate that the dry matter content of skin increased with age. Table 43 shows that the "F" ratio for "sampling dates" was significant at the 1% level. Since, in this experiment, the dry matter values consist of the fat plus protein contents, and since the protein content of the skin remained constant over the experimental period, this increase was due to the increase in the fat component.

TABLE 42.

CHANGES IN THE DRY MATTER CONTENT OF SKIN
EXPRESSED AS % OF THE ORIGINAL WEIGHT.

		<u>Sampling Dates</u>					
		1	2	3	4	5	6
Mean	of 10 lambs	16.0	16.8	17.8	18.7	19.2	20.0
Standard deviation	excluding lambs 2 and 8	0.4	0.4	0.4	0.6	0.5	0.8
Lamb 2**		17.0	16.5	14.0	19.9	19.2	17.5
Lamb 8**		17.5	18.1	18.6	15.6	16.2	19.0
Mean	of 12 lambs	16.2	16.9	17.6	18.6	18.9	19.9

** badly growing lambs.

TABLE 43.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED
IN TABLE 42.

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	1	0.45	< 1	N.S.
Between lambs within Latin squares	10	6.28	2.91	* *
Between positions within Latin squares	10	5.50	2.55	*
Between dates	5	21.26	9.84	* *
Sampling dates x Latin squares	5	3.47	1.61	N.S.
Error	39	2.16		
Total	70			

Mean values for the chemical composition of the skin obtained at each of the six positions are given in Table 35. Table 37 shows that the "F" ratio for "sampling positions" was significant at the 1% level for the fat content values. On the other hand, Table 41 indicates that the "F" ratio for "sampling positions" was non significant for the protein content values. Table 43 shows that the "F" ratio for "sampling positions" was significant at the 5% level for the dry matter content values. As has been pointed out previously, the dry matter values consisted of the fat plus protein values, and since the protein content values showed no positional variation, it was the positional variation in the fat component which was responsible for the positional variation in the dry matter content of the skin.

Since the positional variations in the fat content did not correspond with those in the thickness measurements, it appears unlikely (as suggested previously), that they were due to the presence of a variable amount of subcutaneous fat. Moreover they did not show an orderly gradient over the sampling area. A comparison of the magnitude of the variations between positions found in this experiment, with those described in Chapter IB, suggest that the former might be due to a combination of the errors in the chemical technique and day to day fluctuations in the chemical composition of the skin unrelated to positional trends.

The differences between lambs in skin thickness and chemical composition were found to be significant at the 1% level indicating that animals varied considerably in the absolute values of the skin characteristics studied, though not in the general pattern of the changes occurring during the experimental season.

A similarity of values for protein content of skin obtained by taking the weight of the fat-free dried residue and by the Kjeldahl method observed in the Preliminary Experiments (Chapter IB) was confirmed. A coefficient of correlation was calculated on 100 samples (those used in the Preliminary Experiments and from the experiment described in this chapter) and found to be of the order of +0.943. A regression equation was calculated (Snedecor, 1946):-

$$Y = -0.85 + 1.05 X \quad \text{or}$$

$$Y = 15.23 + 1.05 (X - 15.32)$$

where Y = protein content obtained by the Kjeldahl method.

X = protein content obtained by weighing the dried fat free residue.

This means that in future experiments, the time consuming Kjeldahl estimations could be omitted and the dried fat, free residue assumed to consist entirely of protein.

DISCUSSION.

From the results of this experiment it appears that within 10 days after birth, the skin of a lamb is as thick as at five months of age.

Unfortunately, no measurement was obtained at birth and it is possible therefore, that the high value obtained at the first sampling was the result of a rapid post-natal increase in skin thickness. A significant correlation between skin thickness and age at the first sampling supports this hypothesis. On the other hand, higher correlations exist between skin thickness and liveweight and also between skin thickness and liveweight gain between birth and the first sampling. Since weight and age are strongly correlated at the first sampling, it cannot be ascertained whether older lambs had thicker skins because they were heavier, whether heavier lambs had thicker skins because they were older or possibly, that both skin thickness and liveweight increased over that period without being interdependent.

Between the first and the second or third samplings, skin increased in thickness by approximately 14%. By the fourth sampling it had decreased to the value obtained at the first sampling and, until the end of the experiment at about five months of age, it did not alter in thickness.

It is impossible to determine from the data obtained in this experiment, whether the skin thickness values obtained at the second and third samplings represent the maximum post-natal increase in skin thickness or whether skin thickness reached its maximum value at some point

in time between one and 12 weeks of age. Likewise, the decline in skin thickness which occurred between the third and fourth samplings might have begun at any time between nine and 13 weeks after birth.

A study of the changes in the histological structure of the skin occurring during that period, together with a more precise definition of the shape of the post-natal curve, is necessary before an explanation of these changes can be more than speculative.

Changes in the wool follicle and fibre populations in the Romney breed occurring during the early post-natal period, have been studied by several workers. Burns (1949) found that the initiation of the secondary follicles stopped at approximately three months of age. Her results, however, must be treated with caution because they are based on studies of two lambs only, both with poor growth curves, only one surviving longer than six months after birth. Since moreover, she sampled at three monthly intervals, the period of follicle initiation is ill defined. Similar time relationships were, however, found by Goot (1945). Henderson (1953), working on fibre populations of 12 lambs and sampling at four weeks after birth and at eight weekly intervals thereafter, found that the major increase in fibre numbers took place before 12 weeks of age.

It is possible that both follicle formation and skin thickness are a reflection of the physiological activity of the skin and that a peak of this activity is reached five to 12 weeks after birth.

An alternative explanation of the rise and decline in skin thickness within the first five months after birth could be derived from the results obtained in the Shearing Experiment (Chapter IV), which indicate that after shearing skin increases markedly in thickness. It is possible that skin responds to changes in temperature by changes in thickness and that the increased thickness after shearing is caused by a lowering of the skin temperature following the removal of wool. If this hypothesis is true, then the expulsion of the lamb from the uterus into a colder environment could lead to an increase in skin thickness. Then, as the length of wool increases and the environmental temperature rises with approaching summer, skin thickness falls to a lower value. Again, a more precise definition of the skin thickness curve and of the histological picture, together with a study of the effects of changes in the environmental temperature on skin thickness, might throw some light on this question.

Of interest in connection with the problem of tearing pelts, is the fact that the two lambs which grew more slowly than the rest of the experimental group, had thinner skins throughout and that positive correlations existed between skin thickness and liveweight and also between skin thickness and liveweight gain at each sampling. These results are in agreement with those of Clarke, Stuart and Frey (1937), who found that in a post-weaning study of nine pairs of Rambouillet lambs on widely differing planes of nutrition, the underfed

lambs had skins about 57% of the thickness of the fully fed ones. Their methods of measurement of skin thickness have been criticised in Chapter IA, while conclusions based on the few animals studied in these investigations must necessarily be regarded as tentative, but it would seem that slow growth prior to slaughter due to under-nutrition or other unfavourable environmental conditions, could produce thinner pelts which were liable to tearing. A study of the effects of widely differing planes of nutrition from birth to five months on the thickness, histology and chemical composition of the skin in lambs would elucidate the importance of the nutritional factor.

The protein content of skin remained constant during the entire experimental period, but the fat content rose with age. The skin of the two poorly growing lambs had lower fat values throughout. Their protein values, however, were similar to the ones obtained for the remainder of the experimental group. These results confirm the findings of Clarke, Stuart and Frey (1937), who found that the underfed lambs had skins with a lower fat content. They are also parallel to the results of workers such as Hammond (1932), Wallace (1948) and Palsson and Verges (1952), who found that the fat content of tissues in the carcass of sheep increased with age and was lower in animals which had been underfed prior to slaughter.

SUMMARY.

Changes in the thickness, histology and chemical composition of the skin in sheep have been observed at monthly intervals from within 10 days after birth until five months of age in 12 Romney lambs. Results obtained indicate that:-

1. Skin increases in thickness until the lamb is five to 10 weeks of age. It then decreases to the value obtained at birth and retains this thickness until five months of age.

2. The fat content of skin increases with age, but the protein content remains constant. The total dry matter content increases with age.

3. The skin of the slower growing lambs is thinner and has a lower fat content.

CHAPTER IV

THE SHEARING EXPERIMENT.

INTRODUCTION.

The effect of shearing on the thickness and chemical composition of the skin was studied in 16 Romney ewes.

I. METHODS AND MATERIALS.

(a) Plan of Experiment.

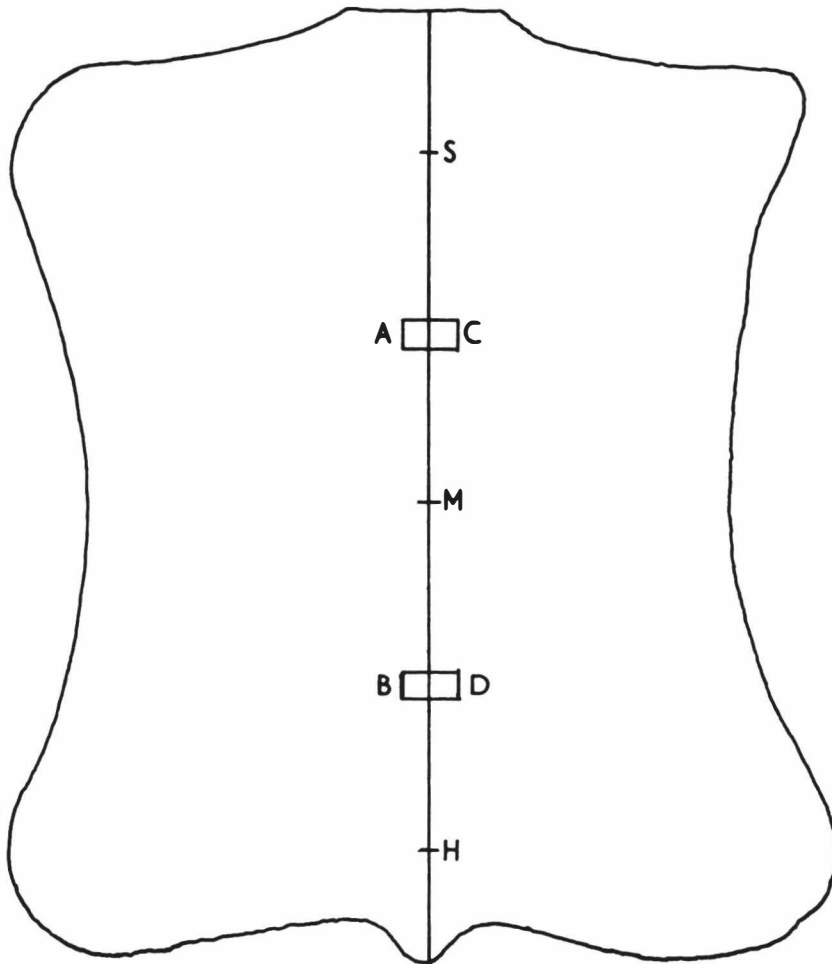
Sixteen Romney ewes were sampled:

1. Two days before shearing, on 6th January, 1953.
2. " " after " on 19th January, 1953.
3. 14 " " " on 22nd January, 1953.
4. 31 " " " on 8th February, 1953.

Since the results described in Chapter III suggested that a weak postero-anterior gradient in skin thickness might exist, the samples for this experiment were taken further away from the anterior and the posterior ends of the sheep and nearer to the middle of the back. To reduce the variations between samples in the distance from the midline which, because of the steep dorso-ventral gradient in skin thickness, might introduce variations between samples other than those caused by the experimental treatment, samples were taken immediately adjacent to the vertebral column. A key to the sampling positions used appears in Diagram 6. However, a Latin square design consisting of four 4 by 4 Latin squares was used (see Table 44) to prevent the possible confounding of the treatment effects by positional

DIAGRAM 6

Sampling positions used in the shearing experiment.



S Vertebral column at the shoulder level.

H Vertebral column at the hip-bone level.

SM=HM

A Sample situated half way between S and M to the left of and adjacent to the vertebral column.

B Sample situated half way between H and M to the left of and adjacent to the vertebral column.

C & D Samples similarly situated to A and B but to the right of the vertebral column.

trends or differences between sheep.

A possible weakness of the design lay in the fact that only one sample was taken prior to shearing. However, the animals were not available until a few days before shearing. Moreover, results from the Skin Development Experiment (Chapter III) indicated that even in growing lambs, skin thickness varied but little over long periods. It was therefore considered, that the assumption that prior to shearing skin thickness remained constant, was valid and that any changes following the first sampling were due to the effect of shearing.

Besides the skin thickness and the chemical composition determinations no other measurements were made. Liveweight measurements were not recorded because sampling took place some distance away from the scales and weighing was not practicable.

TABLE 44.

PLAN OF THE SHEARING EXPERIMENT.

Ewe's Exp. No.	No. of Sampling Date			
	1	2	3	4
1	A	B	C	D
2	B	A	D	C
3	C	D	B	A
4	D	C	A	B

LATIN SQUARE 1

5	A	B	C	D
6	B	C	D	A
7	C	D	A	B
8	D	A	B	C

LATIN SQUARE 2

9	A	B	C	D
10	B	D	A	C
11	C	A	D	B
12	D	C	B	A

LATIN SQUARE 3

13	A	B	C	D
14	B	A	D	C
15	C	D	A	B
16	D	C	B	A

LATIN SQUARE 4

(b) Experimental Animals

Fifteen five-and-a-half year old ewes from Dr. L.R. Wallace's Fertility Flock and one three-and-a-half year old ewe (number 16) from the Ruakura Hill Country Research Station, were used in this experiment. All ewes had reared a lamb and weaning had taken place on the 9th December. All ewes were in good store condition. In spite of the fact that four samples were taken within a period of a month, two of the operations taking place within four days, the wounds healed quickly with no infection and the animals did not appear to be affected by the operation. The list of the sheep used appears in Table 44.

(c) Experimental Technique.

Skin thickness was measured with the Instrument. The samples were shaved after removal from the animal and kept in a refrigerator for 24 hours prior to being measured. Each sample consisted of four equal squares, each square being one square centimetre in area. Each square was measured five times and the mean for the twenty measurements per sample was obtained. The details of the technique used have been described in Sections I (a)(1) and II (b)(4) of Chapter I(A).

Since it was found in Chapter III that a correlation of +0.943 existed between the protein content values obtained by the Kjeldahl method and those obtained by weighing the dried fat free residue, the former method was abandoned and the residue assumed to consist entirely of protein.

II. RESULTS.

(a) Skin Thickness.

The mean values for skin thickness together with the standard deviations are given in Table 45 and Graph 10. Graph 9 shows the individual skin thickness values for each of the 16 sheep. It can be seen that without exception, skin increased markedly in thickness after shearing. Table 46 shows that the differences between "sampling dates" were significant at the 1% level. The "dates x Latin squares" interaction was non significant indicating that there was no differential response in skin thickness between the groups of sheep in the different Latin squares. In Table 47, the increase in skin thickness after shearing, for each of the 16 sheep, is expressed as a percentage of the pre-shearing thickness.

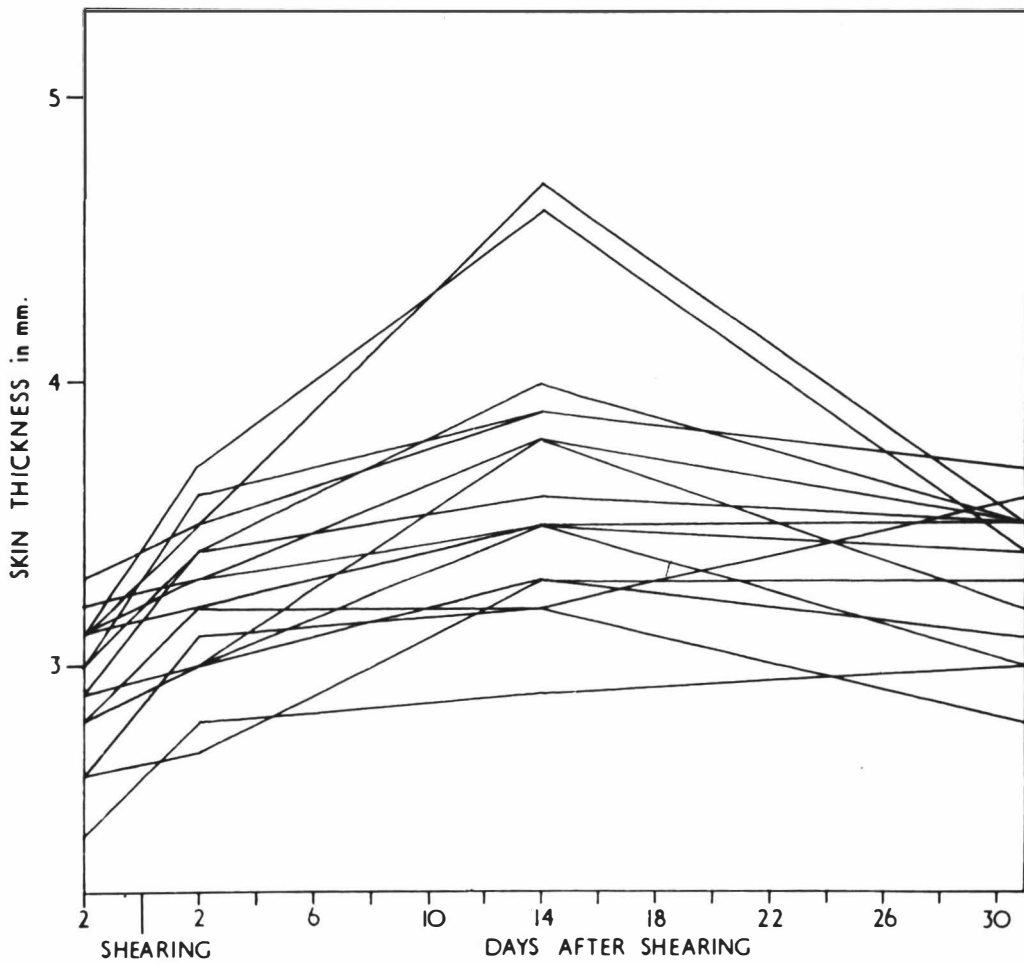
This change in skin thickness was evident at the second sampling (two days after shearing) and was at a maximum at the third sampling (14 days after shearing). At the fourth sampling (31 days after shearing), skin had decreased in thickness as compared to the third sampling but it was still thicker than at the second sampling.

From this data it can be deduced that the change in skin thickness had begun within 48 hours after shearing and that 31 days after shearing skin thickness had already begun to decrease. However, the magnitude of this change is unknown and the peak could lie between three and 30 days after shearing. Moreover, the size,

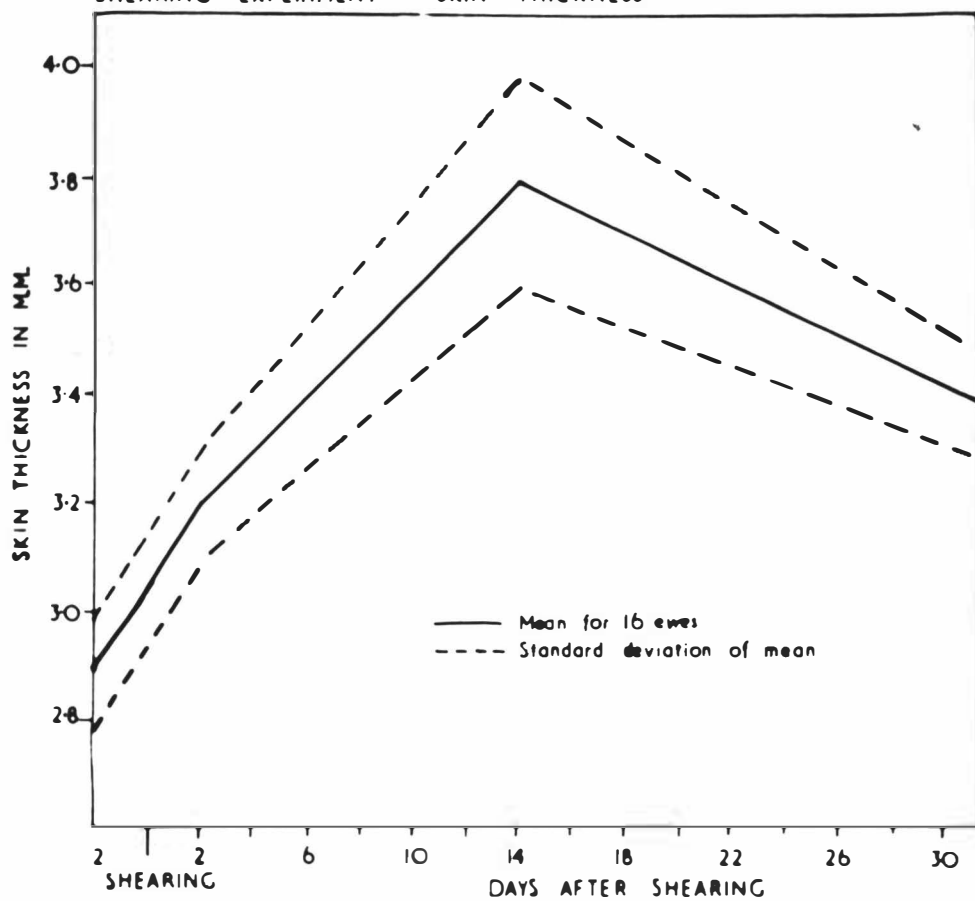
GRAPH 9

SHEARING EXPERIMENT

INDIVIDUAL MEASUREMENTS FOR 16 EWES.



GRAPH 10
SHEARING EXPERIMENT — SKIN THICKNESS



speed and duration of the response could vary between sheep and be influenced by various environmental factors.

Table 46 shows that there was no appreciable difference in skin thickness between positions indicating that the area sampled was uniform in thickness.

Table 47 shows that sheep varied in the magnitude and speed of response to the shearing treatment.

Table 46 shows that the "F" ratio for "sheep" was significant at the 1% level.

A comparison of Graphs 9 and 4 shows that the average skin thickness of the ewes prior to shearing (2.9 millimetres) was similar to that of the 12 lambs sampled within 10 days of birth (2.9 millimetres). Though this is not a valid comparison since the two groups of animals were of different sex, and moreover nothing was known of the past history of the ewes, it suggests that skin does not increase markedly in thickness as the animals grow older.

TABLE 45.

CHANGES IN SKIN THICKNESS DURING THE
SHEARING EXPERIMENT (Millimetres)

	Before Shearing	<u>After Shearing</u>		
		2 days	14 days	31 days
Mean	2.9	3.2	3.8	3.4
Standard deviation	0.1	0.1	0.2	0.1

TABLE 46.

ANALYSIS OF VARIANCE OF THE DATA PRESENTED IN

TABLE 45.

(For convenience all calculations for the Analysis of Variance in this table are in tenths of a millimetre.)

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	3	281.3	4.06	*
Between sheep within Latin squares	12	435.6	6.30	**
Between positions within Latin squares.	12	64.46	< 1	N.S.
Between dates	3	1,837.9	26.59	**
Dates x Latin squares	9	122.23	1.77	N.S.
Error	24	69.12		
Total	70			

TABLE 47.

INCREASE IN SKIN THICKNESS AFTER SHEARING
EXPRESSED AS A PERCENTAGE OF THE PRE-SHEARING THICK-
NESS.

Sheep No.	2 days after shearing	14 days after shearing	31 days after shearing
1	3.4	31.0	20.7
2	19.2	23.1	7.7
3	7.1	17.8	19.7
4	14.3	25.0	25.0
5	6.1	42.4	1.4
6	19.3	80.6	15.1
7	3.4	20.7	3.4
8	16.6	20.8	25.0
9	17.2	37.9	20.7
10	3.8	26.9	26.9
11	13.3	30.0	23.3
12	13.3	20.0	20.0
13	12.9	25.8	19.3
14	3.1	18.7	0
15	3.2	22.6	16.1
16	6.5	12.9	9.7
Mean	10.2	28.5	15.9

(b) Chemical Composition.

Tables 49 and 50 show that there was no significant difference in the fat and protein contents of the skin between sampling dates. Thus the change in skin thickness following shearing was unaccompanied by a change in the chemical composition of the skin.

There was no significant difference between positions in either the fat or the protein content of the skin. However, there was a difference significant at the 1% level between sheep in both the fat and the protein contents of the skin, indicating a large variation between sheep in the chemical composition of skin.

All the percentages were analysed as such because they did not appear to violate the conditions required for the Analysis of Variance, outlined in Section II, Chapter III. Bartlett's test in particular revealed a homogeneity of variance.

TABLE 48.

ANALYSIS OF VARIANCE OF THE EFFECT OF SHEARING
ON THE FAT CONTENT OF SKIN EXPRESSED AS % OF THE
ORIGINAL WEIGHT.

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	3	45.36	18.44	* *
Between sheep within Latin squares	12	48.53	19.73	* *
Between positions within Latin squares	12	2.80	1.14	N.S.
Between dates within Latin squares	12	5.33	2.17	N.S.
Error	24	2.46		
Total	63			

TABLE 49.

ANALYSIS OF VARIANCE OF THE EFFECT OF SHEARING ON
THE PROTEIN CONTENT OF SKIN EXPRESSED AS
% OF THE ORIGINAL WEIGHT.

Source of Variation	d.f.	M.S.	F.	Level of Significance
Between Latin squares	3	8.95	4.81	* *
Between sheep within Latin squares	12	7.89	4.24	* *
Between positions within Latin squares	12	1.69	< 1	N.S.
Between dates within Latin squares	12	1.74	< 1	N.S.
Error	24	1.86		
Total	63			

DISCUSSION.

From the results of this experiment it appears that skin increases in thickness after shearing, this increase being unaccompanied by changes in the chemical composition of the skin. Because of the limited number of samplings, the exact magnitude, speed and duration of this response are unknown, but the change was evident 48 hours after shearing and past its maximum value 31 days after shearing. It is not known whether the nature and the extent of the response can be influenced by environmental factors such as plane of nutrition and temperature.

Evidence for an effect of shearing on the rate of wool growth is not clear because other experimental treatments tend to confound the possible effects of shearing. Ferguson, Carter and Hardy (1949) found a positive

correlation between the rate of wool growth and environmental temperature. However, examination of their data shows that an increase in wool growth occurred immediately after shearing. On the other hand, Coop (1953) did not find an increase in the rate of wool growth after shearing, although his results may be confounded, partly by changes in liveweight during the experimental period and partly by a superimposed light treatment.

Cockrem (1952) working with four Romney N-type sheep (see Dry, (1935)) and using no unshorn controls, found that within 24 hours after shearing, rectal temperatures fell (by about one degree Fahrenheit) and pulse rates increased (by an average of 26 beats per minute). If this is a normal post-shearing effect, it is possible that it causes a dilation of the peripheral blood vessels leading to an increase in the physiological activity of the skin which in turn results in the development of new cell layers and an increase in skin thickness. This response could form a part of the temperature regulating mechanism of the sheep. Thus it is possible that skin responds to changes in temperature by changes in thickness and that the increased thickness after shearing is caused by a lowering of the skin temperature following the removal of wool.

A study of the changes in the histological structure of the skin after shearing, a more precise definition of the post shearing changes in skin thickness and information on the effect/^{of} environmental temperature on skin thickness, seem necessary before any theory explaining the increase in skin thickness after shearing can be more than speculative. An experiment in which shorn and unshorn groups

of sheep are submitted to different environmental temperatures, might supply an answer to this question.

In connection with the problem of the tearing of lamb pelts, it should be noted that if the skin of lambs also increases in thickness after shearing, unshorn lambs would tend to have thinner pelts than recently shorn ones.

Lastly, it can be seen that skin thickness values of the ewes prior to shearing are similar to those of lambs sampled within 10 days after birth. This comparison is of doubtful value because the two groups of sheep are of different sex and moreover, the previous history of the ewes is unknown, but it would seem that skin does not increase markedly in thickness in the adult sheep.

SUMMARY.

Changes in the thickness, histology and chemical composition of the skin following shearing were studied in 16 Romney ewes. Results obtained indicate that:-

1. Skin increases in thickness after shearing, although because of limited number of samplings the exact magnitude, speed and duration of response are unknown.

2. The chemical composition of the skin in sheep remains constant after shearing.

GENERAL SUMMARY AND DISCUSSION

In the course of these investigations, an instrument has been developed for the rapid and accurate measurement of skin thickness while techniques have been adapted for the determination of the fat and protein contents of the skin in sheep. The distribution of gradients in skin thickness over the body of a sheep has been determined and a uniform sampling area defined. Changes in the thickness and chemical composition of the skin have been followed from birth to five months in lambs and also after shearing, in adult sheep.

Results of these experiments have been discussed at the end of each chapter, but in connection with the problem of the tearing of lamb pelts it has been observed that skins of lambs with slower growth rates are thinner than those of more rapidly growing animals. It is also probable that unshorn lambs have thinner skins than lambs which have been shorn recently.

In addition, some points of general interest arise from the results of these experiments. Firstly, the large differences between sheep in the characteristics studied, emphasize the usefulness of an experimental approach where the same animal is studied over a period. In experiments where groups of animals are slaughtered at several points in time, although a more complete set of measurements can be obtained for each group at a given point in time, changes occurring between two slaughter dates may be obscured by large variations

between animals.

Secondly, these investigations have also emphasized the scarcity of existing knowledge of the histological and physiological changes (other than those connected with the wool follicle and fibre) occurring in the skin of sheep during growth, and later in adult life. This knowledge is essential if conditions causing skin weaknesses and leading to pelt and leather defects are to be understood and, if possible, eliminated.

Leather characteristics are the results of both the nature of the original product and the fellmongering and tanning processes. It is improvements in the quality of the original product, skin, which lie within the province of the agriculturalist. With this aim in mind, knowledge of the histology, physiology and biochemistry of the skin should be extended. Changes occurring in the skin structure and chemical composition during the fellmongering and tanning processes should be studied in order that those skin characteristics which influence leather quality may be determined. At the same time, a survey of the problems of the pelt and leather industries and classification of defects into those caused by inherent skin weaknesses and those resulting from faulty processing, would help to define problems of economic importance which await investigation.

REFERENCES.

- AUBER, L. (1950): Trans. Roy. Soc. Edin. LXII: 191
- BRODY, S. (1945): Bioenergetics and Growth. New York, Reinhold Publishing Corporation.
- BURNS, M. (1949): J. Agric. Sci. 39: 64.
- CARTER, H.B. (1943): C.S.I.R.O. Bull. 164.
- CARTER, H.B. and HARDY, M. (1947): C.S.I.R.O. Bull: 215.
- CARSTENS, P. and KINZELBACH, W. (1943):
Zuchts. 8: 135.
- CLARKE, T.D., STUART, L.S. and FREY, R.W. (1937):
Sonderdruck aus der Stiasny
Festschrift, 144.
- COCKREM, F.R. (1952): Private Communication.
- COOP, I.E. (1953): J. Agric. Sci. 43: 456.
- DRY, F.W. (1935): N.Z.J. Agric. Sci. 51: 229.
- FERGUSON, K.A.; CARTER, H.B. and HARDY, M. (1949):
Aust. J.Sci. Res. B2: 42.
- FISHER, R.A. (1925): The Design of Experiments.
Oliver & Boyd, Edinburgh.
- FISHER, R.A. and YATES, F. (1953): Statistical Tables.
Oliver & Boyd, Edinburgh.
- GALPIN, N. (1947): J. Agric. Sci. 37: 275.
- GALPIN, N. (1948): J. Agric. Sci. 38: 303.
- GOOT, H. (1945) : N.Z.J. Sci. Tech. 27: 173.
- HAMMOND, J. (1932): Growth and Development of Mutton
Qualities in the Sheep.
Oliver Boyd, Edinburgh.
- HENDERSON, A.E. (1953): J. Agric. Sci. 43: 12.
- McMAHON, P.R. (1937): J. Text. Inst. XXVIII: T349.
- NICOV, T. (1931): Zeitschr. f. Zucht. B.XXI: 351.
- PALSSON, H. and VERGES, J.B. (1952): J. Agric. Sci. 42: 1.
- RYDER, M.L. (1953): Wool Ind. Res. Ass'n. Bull. 15: 104.
- SCHINCKEL, P.G. (1953): Nature 171: 310.
- SNEDECOR, G.W. (1946): Statistical Methods.
Iowa State College Press.
- WALLACE, L.R. (1948): J. Agric. Sci. 38: 93, 243, 367.
-

APPENDIX A

DEVELOPMENT OF TECHNIQUES

TABLE I

THE EFFECT ON SKIN THICKNESS OF SHAVING BEFORE
OR AFTER REMOVAL FROM THE ANIMAL

(Instrument Measurements in Millimetres)

Shaven Before				Shaven After			
<u>Lot</u>	<u>Piece</u>	<u>Square</u>	<u>Mean</u>	<u>Lot</u>	<u>Piece</u>	<u>Square</u>	<u>Mean</u>
I	1	1	2.5	I	1A	1	2.5
		2	2.6			2	2.4
		3	2.5			3	2.5
		4	2.5			4	2.5
		5	2.5			5	2.4
		6	2.6			6	2.6
	Mean		2.5			2.5	
II	2	1	3.0	II	2A	1	2.8
		2	2.8			2	2.9
		3	3.0			3	3.0
		4	2.9			4	2.9
		5	3.1			5	2.9
		6	2.9			6	2.8
	Mean		2.9			2.9	
III	3	1	3.1	III	3A	1	3.1
		2	3.1			2	3.3
		3	3.0			3	3.1
		4	3.0			4	3.1
		5	3.0			5	3.1
		6	2.9			6	3.1
	Mean		3.0			3.1	
IV	4	1	2.0	IV	4A	1	2.1
		2	2.3			2	2.2
		3	2.1			3	1.9
		4	2.4			4	2.3
		5	2.2			5	2.4
		6	2.4			6	2.3
	Mean		2.2			2.2	

TABLE II

THE EFFECT ON SKIN THICKNESS OF STORAGE IN A
DEEP FREEZE FOR 24 HOURS

(Instrument Measurements in Millimetres)

<u>Lot</u>	<u>Piece</u>	<u>Square</u>	Fresh	After deep freeze
			<u>Mean</u>	<u>Mean</u>
I	1	1	2.5	2.8
		2	2.6	2.8
		3	2.5	2.8
		4	2.5	2.7
		5	2.5	2.8
		6	2.6	2.8
		Mean		2.5
I	1A	1	2.5	2.6
		2	2.4	2.6
		3	2.5	2.5
		4	2.5	2.5
		5	2.4	2.5
		6	2.6	2.6
		Mean		2.5
II	2	1	3.0	3.0
		2	2.8	3.0
		3	3.0	3.1
		4	2.9	3.1
		5	3.1	3.1
		6	2.9	3.1
		Mean		2.9
II	2A	1	2.8	3.0
		2	2.9	3.0
		3	3.0	3.0
		4	2.9	3.1
		5	2.9	3.1
		6	2.8	3.1
		Mean		2.9

TABLE III

THE EFFECT ON SKIN THICKNESS OF STORAGE IN
A REFRIGERATOR FOR 24 HOURS.

(Instrument Measurements in Millimetres)

<u>Lot</u>	<u>Piece</u>	<u>Square</u>	<u>Fresh</u> <u>Mean</u>	<u>After refrigerator</u> <u>Mean</u>
III	3	1	3.1	3.0
		2	3.1	3.0
		3	3.0	3.0
		4	3.0	3.0
		5	3.0	3.1
		6	2.9	2.9
	Mean		3.0	3.0
III	3A	1	3.1	3.2
		2	3.3	3.3
		3	3.1	3.2
		4	3.1	3.1
		5	3.1	3.2
		6	3.1	3.1
	Mean		3.1	3.2
IV	4	1	2.0	2.0
		2	2.3	2.3
		3	2.1	2.1
		4	2.4	2.3
		5	2.2	2.2
		6	2.4	2.4
	Mean		2.2	2.2
IV	4A	1	2.1	2.1
		2	2.2	2.2
		3	1.9	2.0
		4	2.3	2.3
		5	2.4	2.4
		6	2.3	2.3
	Mean		2.2	2.2

TABLE IV

THE EFFECT ON SKIN THICKNESS OF SHAVING BEFORE
OR AFTER REMOVAL FROM THE
ANIMAL

(Instrument Measurements in Millimetres)

Shaven Before			Shaven After		
<u>Piece</u>	<u>Square</u>	<u>Mean</u>	<u>Piece</u>	<u>Square</u>	<u>Mean</u>
1	1	1.8	3	1	1.9
	2	1.9		2	1.9
	3	1.8		3	1.9
	4	1.8		4	1.9
Mean		1.8			1.9
2	1	1.9	4	1	1.9
	2	1.9		2	1.9
	3	2.0		3	2.1
	4	2.0		4	2.0
Mean		1.9			2.0

TABLE V

THE EFFECT ON SKIN THICKNESS OF STORAGE IN

THE REFRIGERATOR FOR 24 HOURS

(Instrument Measurements in Millimetres)

<u>Piece</u>	<u>Square</u>	<u>Fresh</u> <u>Mean</u>	<u>After refrigerator</u> <u>Mean</u>
1	1	1.8	1.9
	2	1.9	1.8
	3	1.8	1.9
	4	1.8	1.9
	Mean		1.8
2	1	1.9	1.8
	2	1.9	1.9
	3	2.0	2.0
	4	2.0	2.1
	Mean		1.9
3	1	1.9	1.9
	2	1.9	1.8
	3	1.9	1.9
	4	1.9	1.8
	Mean		1.9
4	1	1.9	1.9
	2	1.9	1.9
	3	2.1	2.1
	4	2.0	2.1
	Mean		2.0

APPENDIX B

THE SKIN DEVELOPMENT EXPERIMENT

TABLE I

SKIN THICKNESS

(Instrument Measurements in
Millimetres)

Lamb	<u>Sampling Date</u>						Mean
	1	2	3	4	5	6	
1	3.6	3.3	3.5	3.0	3.0	3.4	3.3
2 **	2.7	2.1	2.5	2.6	2.7	2.6	2.5
3	2.9	3.0	3.3	3.1	2.6	3.0	3.0
4	2.7	2.9	3.4	3.4	2.5	2.9	3.0
5	2.6	3.0	3.4	3.0	2.9	2.9	3.0
6	3.4	3.5	2.5	3.0	3.0	3.0	3.1
7	3.1	3.4	3.2	2.9	2.8	2.8	3.0
8 **	2.8	2.9	3.1	2.5	2.5	2.5	2.7
9	2.1	3.7	3.6	2.7	2.8	2.8	2.9
10	2.1	3.3	3.4	3.1	3.5	3.0	3.1
11	3.4	4.2	3.3	3.2	3.1	*	2.9
12	2.6	2.8	3.1	2.8	2.5	2.5	2.7
Mean	2.8	3.2	3.2	2.9	2.8	2.8	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE IIWEIGHTS OF LAMBS IN POUNDS

Lamb	Birth Weight	Sampling Date						Weaning*** Weight
		1	2	3	4	5	6	
1	12	18	37	53	69	71	84	61
2	8	13	21	32	39	43	54	38
3	9	13	30	46	61	64	74	61
4	8	12	35	50	65	68	83	66
5	8	12	33	49	64	61	72	63
6	11	17	36	49	60	70	84	65
7	10	15	36	52	62	70	72	61
8	9	13	18	32	39	47	50	41
9	7	11	33	50	58	62	65	59
10	7	11	32	49	61	70	73	63
11	12	19	39	55	67	75	*	63
12	9	13	34	53	63	69	70	64
Mean		14	32	47	59	64	71	59

* died , suspected cause pulpy kidney.

** suffering from a septic eye.

*** weaning weights taken on 9.12.53 i.e. 19 days after the fourth sampling for lambs 1-5 inclusive, 9 days after the fourth sampling for lambs 7-12 inclusive, and 9 days after the third sampling for lamb 6.

TABLE III

HEIGHT AT WITHERS

(Centimetres)

Lamb	<u>Sampling date</u>					
	1	2	3	4	5	6
1	33	42	45	47	48	53
2**	32	37	38	43	46	48
3	34	38	48	51	53	56
4	36	39	46	48	51	53
5	34	40	46	51	43	52
6	37	45	47	48	54	56
7	35	42	46	48	51	54
8**	36	37	45	48	49	52
9	35	39	45	47	48	50
10	38	39	44	49	51	54
11	37	43	49	50	53	*
12	37	39	46	48	51	54
Mean	35	40	45	48	50	53

TABLE IV

STAPLE LENGTH
(Centimetres)

Lamb	Sampling date					
	1	2	3	4	5	6
1	0	3	5	7	9	9.5
2**	0	2	3.5	5.5	6	8
3	0	2	5	6	6.5	9
4	0	2.5	4	7.5	10	11
5	0	2.5	4	6	8.5	9
6	0	3.5	6	9	10	11
7	0	3	4	6	7	11
8**	0	3	5.5	7	8.5	9
9	0	2.5	5	6.5	7	8
10	0	2.5	4	6	7	11
11	0	2.5	6	7.5	9.5	*
12	0	2.5	6	7.5	8	8
Mean		2.6	4.8	6.8	8.1	9.5

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE V.

FAT CONTENT EXPRESSED AS % OF ORIGINAL
WEIGHT

Lamb	<u>Sampling date</u>						Mean
	1	2	3	4	5	6	
1	2.4	3.1	2.4	3.3	3.9	4.1	3.2
2**	2.2	1.8	2.5	3.6	5.4	2.7	3.0
3	1.6	2.7	2.0	5.8	6.1	6.3	4.1
4	1.6	3.2	4.2	5.8	8.0	8.4	5.2
5	1.6	2.8	2.9	3.8	3.7	3.8	3.1
6	2.6	1.7	3.0	2.5	4.1	3.6	2.9
7	2.4	4.6	5.1	4.7	5.3	6.1	4.7
8* *	1.8	2.7	2.1	2.0	2.0	2.1	2.1
9	1.7	2.7	2.5	2.6	1.8	4.0	2.5
10	1.5	2.8	2.8	4.1	5.6	4.1	3.5
11	2.1	3.8	3.5	5.0	6.8	*	4.2
12	1.9	2.7	3.7	7.7	6.4	9.6	5.3
Mean	1.9	2.9	3.0	4.2	4.9	5.0	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE VI.

FAT CONTENT EXPRESSED AS % OF DRY MATTER

Lamb	<u>Sampling date</u>						Mean
	1	2	3	4	5	6	
1	16.5	18.2	15.0	18.9	23.6	24.7	19.5
2**	12.8	11.0	17.8	18.0	28.2	15.4	17.2
3	10.3	15.3	11.5	28.6	31.4	28.5	20.9
4	10.6	17.6	22.4	27.1	37.0	37.5	25.4
5	10.6	15.9	15.5	19.7	19.1	19.2	16.7
6	16.9	10.9	17.7	14.5	19.3	22.0	16.9
7	13.2	24.9	24.7	24.8	28.0	34.3	25.0
8**	10.5	14.8	11.5	12.8	12.4	10.9	12.1
9	9.5	16.0	13.6	13.5	11.2	21.1	14.1
10	10.2	17.1	17.5	24.1	27.4	21.0	19.5
11	13.1	23.9	20.5	33.7	35.6	*	25.4
12	11.5	19.4	20.8	36.2	33.6	38.0	26.6
Mean	12.1	17.1	17.4	22.6	25.6	24.8	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE VII

PROTEIN CONTENT EXPRESSED AS % OF
ORIGINAL WEIGHT SAMPLING
DATE

Lamb	<u>Sampling Date</u>						Mean
	1	2	3	4	5	6	
1	12.2	13.8	13.6	14.1	12.6	12.5	13.1
2**	14.8	14.7	11.5	16.3	13.8	14.8	14.3
3	14.3	15.0	15.7	14.5	13.4	15.7	14.8
4	13.9	15.0	14.7	15.4	13.6	14.1	14.4
5	13.6	14.8	16.1	15.6	15.7	15.8	15.3
6	12.9	14.2	13.9	14.8	17.0	16.5	14.9
7	15.9	14.0	15.6	14.3	13.5	11.8	14.2
8**	15.7	15.4	16.5	13.6	14.2	16.9	15.4
9	15.9	14.0	15.9	16.8	14.4	15.0	15.3
10	13.1	13.5	13.0	13.0	14.9	15.4	13.8
11	14.0	12.3	13.6	9.9	12.3	*	12.4
12	14.8	11.2	14.0	13.6	12.6	15.7	13.6
Mean	14.2	14.0	14.5	14.3	14.0	14.9	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE VIII

PROTEIN CONTENT EXPRESSED AS % OF DRY
MATTER

Lamb	Sampling Date						Mean
	1	2	3	4	5	6	
1	83.5	81.8	85.0	81.1	76.4	75.3	80.5
2**	87.2	89.0	82.2	82.0	71.8	84.6	82.8
3	89.7	84.7	88.5	71.4	68.6	71.5	79.1
4	89.4	82.4	77.6	72.9	63.0	62.5	74.6
5	89.4	84.1	84.5	80.3	80.9	80.8	83.1
6	83.1	89.1	82.3	85.5	80.7	78.0	83.1
7	86.8	75.1	75.3	75.2	72.0	65.7	75.0
8**	89.5	85.2	88.5	87.2	87.6	89.1	87.8
9	90.5	84.0	86.4	86.5	88.8	78.9	85.8
10	89.8	82.9	82.5	75.9	72.6	79.0	80.4
11	86.9	76.1	79.5	66.3	64.4	*	74.6
12	88.5	80.6	79.2	63.8	66.4	62.0	73.4
Mean	87.8	82.9	82.6	77.3	74.4	75.2	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE IX

DRY MATTER CONTENT EXPRESSED AS % OF
ORIGINAL WEIGHT.

Lamb	Sampling Date						Mean
	1	2	3	4	5	6	
1	14.6	17.0	16.0	17.4	16.5	16.6	16.3
2**	17.0	16.5	14.0	19.9	19.2	17.5	17.3
3	15.9	17.7	17.7	20.3	19.5	22.0	18.8
4	15.5	18.2	18.9	21.2	21.6	22.5	19.6
5	15.2	17.6	19.0	19.4	19.4	19.6	18.4
6	15.5	15.9	16.9	17.3	21.0	20.1	17.8
7	18.3	18.6	20.7	19.0	18.8	17.9	18.9
8**	17.5	18.1	18.6	15.6	16.2	19.0	17.5
9	17.6	16.7	18.4	19.4	16.2	19.0	17.9
10	14.6	16.3	15.8	17.1	20.5	19.5	17.3
11	16.1	16.1	17.1	14.9	19.1	*	16.7
12	16.7	13.9	17.7	21.3	19.0	25.3	19.0
Mean	16.2	16.9	17.6	18.6	18.9	19.9	

* died, suspected cause pulpy kidney

** badly growing lamb

TABLE X

PROTEIN CONTENT EXPRESSED AS % OF ORIGINAL
WEIGHT OBTAINED BY THE KJELDAHL METHOD AND BY
WEIGHING THE DRIED FAT FREE RESIDUE

Sample Number	Protein % Residue	Kjeldahl	Sample Number	Protein % Residue	Kjeldahl
1'	16.8	16.6	11	12.2	13.2
2'	16.5	16.7	12	13.8	14.9
3'	13.7	14.7	13	13.6	11.8
4'	15.6	15.8	14	14.1	15.5
5'	14.5	14.4	15	12.6	13.1
6'	17.1	16.9	16	12.5	11.9
7'	18.2	18.8	21	14.8	15.2
8'	25.9	23.4	22	14.7	14.9
9'	22.8	22.8	23	11.5	11.4
10'	23.8	22.0	24	16.3	13.8
11'	24.7	22.8	25	13.8	14.2
1	15.8	15.9	26	14.8	13.9
1A	17.5	17.1	31	14.3	14.8
2A	14.7	14.5	32	15.0	14.9
3A	14.6	15.0	33	15.7	16.1
3	16.7	16.2	34	14.5	13.3
3A	16.2	16.1	35	13.4	13.4
A	16.0	16.5	36	15.7	13.5
A1	16.3	17.0	41	13.9	14.4
B1	16.4	16.6	42	15.0	15.4
B1	15.9	15.7	43	14.7	15.6
C1	17.9	17.1	44	15.4	17.3
C1	15.9	16.5	45	13.6	13.8
D	16.3	16.4	46	14.1	15.4
D1	17.2	16.8	51	13.6	13.7
E	17.0	17.4	52	14.8	14.9
E1	17.5	17.5	53	16.1	17.0
F1	17.0	17.3	54	15.6	17.2
F1	17.7	17.6	55	15.7	15.8

TABLE X (Cont'd)

Sample Number	Protein %		Sample Number	Protein %	
	Residue	Kjeldahl		Residue	Kjeldahl
56	15.8	14.7	92	14.0	14.3
61	12.9	13.2	93	15.9	16.0
62	14.2	14.8	94	16.8	16.7
63	13.9	13.5	95	14.4	15.4
64	14.8	15.1	96	15.0	14.4
65	17.0	16.4	101	13.1	13.0
66	15.9	15.8	102	13.5	14.3
71	15.9	16.7	103	13.0	14.5
72	14.0	14.9	104	13.0	13.0
73	15.6	15.7	105	14.9	15.1
74	14.3	13.9	106	15.4	15.2
75	13.5	13.5	111	14.0	14.7
76	11.8	11.2	112	12.3	13.1
81	15.7	16.3	113	13.6	14.2
82	15.4	16.1	114	9.9	9.8
83	16.5	15.2	115	12.3	12.6
84	13.6	13.2	116 *		
85	14.2	14.5	121	14.8	14.6
86	16.9	16.7	122	11.2	12.5
91	15.9	16.1	123	14.0	14.9
			124	13.6	15.3
			125	12.6	13.0
			126	15.7	15.1

* died, suspected cause pulpy kidney.

APPENDIX C

SHEARING EXPERIMENT

TABLE I.

FAT AND PROTEIN CONTENT EXPRESSED AS % OF
THE ORIGINAL WEIGHT

	1		2		3		4	
Sheep	fat	protein	fat	protein	fat	protein	fat	protein
1	6.4	16.2	4.5	17.1	4.6	16.7	4.8	17.8
2	3.3	19.5	3.8	16.6	3.6	21.2	2.8	18.2
3	2.6	18.6	3.1	18.0	2.5	17.6	2.4	18.4
4	7.4	14.4	4.9	14.8	5.8	15.1	6.0	15.2
5	6.0	16.4	3.4	18.4	5.9	16.2	5.3	17.6
6	8.0	16.8	8.7	16.9	16.7	14.1	8.8	14.4
7	3.0	19.1	1.9	17.0	3.1	19.3	2.1	18.1
8	4.7	20.2	2.2	12.7	3.5	18.3	3.6	17.3
9	6.5	12.4	7.0	17.0	9.0	16.8	7.2	13.3
10	3.4	18.5	3.3	18.1	2.9	17.6	2.9	16.9
11	11.1	17.4	7.5	16.0	8.4	18.1	10.4	16.6
12	11.3	14.2	12.1	14.5	12.2	14.9	8.9	14.6
13	6.5	16.5	5.3	15.7	5.6	17.3	4.2	16.6
14	16.8	14.6	12.6	13.7	15.7	14.3	8.6	14.7
15	13.3	15.1	6.8	14.9	7.7	16.6	7.1	15.8
16	2.7	16.6	2.6	14.7	2.5	17.2	2.8	16.6

TABLE II.

FAT AND PROTEIN CONTENT, AND THE CHANGES IN THE
THICKNESS OF THE SKIN DURING THE SHEARING
EXPERIMENT

Sheep	Expressed as % of the original weight		Skin Thickness Before Shearing (mm.)	Increase in skin thick- ness expressed as % of the pre-shearing thickness		
	Mean Fat Content	Mean Protein Content		2 days after shearing	14 days after shearing	31 days after shearing
1	5.1	16.9	2.9	3.4	31.0	20.7
2	3.4	18.9	2.6	19.2	23.1	7.7
3	2.6	18.1	2.8	7.1	17.8	19.7
4	6.0	14.9	2.8	14.3	25.0	25.0
5	5.1	17.1	3.3	6.1	42.4	1.4
6	10.5	15.5	3.1	19.3	80.6	16.1
7	2.5	18.4	2.9	3.4	20.7	3.4
8	3.5	17.1	2.4	16.6	20.8	25.0
9	7.4	14.9	2.9	17.2	37.9	20.7
10	3.1	17.8	2.6	3.8	26.9	26.9
11	9.3	17.0	3.0	13.3	30.0	23.3
12	11.1	14.5	3.0	13.3	20.0	20.0
13	5.4	16.5	3.1	12.9	25.8	19.3
14	13.4	14.3	3.2	3.1	18.7	0
15	8.7	15.6	3.1	3.2	22.6	16.1
16	2.6	16.3	3.1	6.5	12.9	9.7