

THE SPATIAL RELATIONSHIPS BETWEEN SKIN FOLLICLES DURING THEIR DEVELOPMENT IN SHEEP

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Summary

The spatial relationships among primary skin follicles and among the total skin follicle population have been measured and compared during follicle initiation and development in foetuses of Romney \times Southdown cross and Merino sheep.

There was no evidence suggesting that the distribution of primary follicles is different between the two breeds and the number of secondaries that develop is evidently independent of any influence the pattern of primary follicles may have on the pattern of secondary follicle initiation.

After primary follicle initiation is complete, movement of lateral primaries away from their corresponding central primary is greater than that resulting from a uniform expansion of the skin.

After the commencement of secondary initiation changes in the distribution of all follicles are dependent on the S/P ratio. The tendency for an aggregated follicle distribution to result from localized secondary initiation between the primaries in each follicle group is to a large extent counter-balanced by a differential movement of the secondaries away from the primaries.

I. INTRODUCTION

Differences between primary and secondary skin follicles and between the fibres they produce in sheep have led to the conclusion that the relative numbers of these follicle types are of major importance in the determination of fleece structure (Duerden 1939; Wildman and Carter 1939; Carter 1942; Fraser 1953; Carter and Clarke 1957*a*, 1957*b*). Apart from morphological differences, the characterization of primary and secondary follicles is twofold. Firstly they occupy consistent relative places in the sequence of follicle initiation and secondly, because the construction of each follicle group proceeds in the same ordered series of steps, the arrangement of follicle types within each group presents a regular pattern. Thus there is a general relationship between the age of a follicle and its position in the follicle group.

The follicle group is recognized as the basic fleece-producing unit in the skin (Duerden 1939; Wildman and Carter 1939; Carter 1940, 1942, 1943) and although its development has been described in a wide range of sheep breeds (see Fraser and Short 1960) the influences which guide its systematic construction are largely unknown. Because the lateral primary follicles are initiated on either side of and in close association with each central primary follicle (Carter 1940) and because, with the appearance of laterals, the foundation of the ultimate group is laid (Carter 1943, 1955), it is possible that the position of laterals is controlled by the existing central primaries. Burns (1953) has suggested that secondary follicles may possibly arise as the result of "induction" by each primary follicle while Carter (1943) regards the areas in which

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secondary initiation occurs to be definite predetermined regions of local activity corresponding with the "wedges of primordial fibro-genetic tissue" described by Duerden (1939). Such a sequence of inductive or predetermining influences arising from the follicles themselves may be compared with the system of development described by Stern (1954) in which development is regarded as "... a series of pre-patterns, each one being a realized pattern as compared to its predecessor and a new pre-pattern as the basis for its successor."

Another aspect of importance concerns the distribution of the follicle groups themselves. Because the location of each group is essentially determined by the central primary follicles this would appear to be a problem related to the patterning among central primaries. Ryder (1956) considered that follicle development was associated with general body development and Stephenson (1959) has shown skin expansion to be the major factor affecting primary follicle initiation. Schinckel (1955), Stephenson (1959), and Schinckel and Short (1961) have concluded that heavier foetuses have a greater total number of primary follicles. The distribution of central primary follicles is highly uniform yet there is no evidence that the pattern has a directional component (Claxton 1962). These considerations suggest that the mechanism controlling the distribution of central primary follicles may be a competitive one whereby new follicles appear at sites most distant from existing follicles. This would correspond with the model proposed by Wigglesworth (1953) to explain the uniform pattern of abdominal sensory hairs in the hemipteron *Rhodnius prolixus*.

Analyses of the spatial relationships among follicles and the changes in these relationships as follicle initiation and development proceed have been made with the object of providing a more solid basis for an understanding of the way in which the follicle groups are built. Further, a sequence of differentiation controlled in the fashion outlined above could result in a cause-and-effect relationship between the patterns of follicles during their initiation and the final group structure.

The suggestion has also been made that follicles may compete with one another on a spatial basis either for follicle-forming or for fibre-forming substrate (Fraser and Short 1952, 1960). Therefore the spatial relationships between follicles could be important from the aspect of spatial competition.

The spatial relationships between central primary follicles during their initiation and development were reported in an earlier paper (Claxton 1962). The results showed that the pattern of central primaries was similar in six body regions of both Romney \times Southdown cross and Merino sheep, and that it was invariant during initiation and subsequent foetal development. The conclusion drawn from this is that the differences in fleece structure between the two sheep breeds are not influenced or conditioned by the arrangement of central primary follicles.

The work reported in the present paper is the second part of an investigation designed to quantitatively analyse and compare the spatial relationships among wool follicles during initiation and development. It deals with the patterns among primary and among primary and secondary follicles during the period from 70 to 140 days of foetal age. The implications of this work with respect to the determination of follicle groups will be discussed in detail elsewhere.

II. MATERIALS AND METHODS

(a) *Experimental Material*

The foetuses were from 6-year-old fine-woolled Australian Merino ewes and 5-year-old Romney ewes. The Merino ewes were mated with a Merino ram of the same strain while the Romney ewes were mated with a Southdown ram. Foetuses were obtained at 10-day intervals from 70 to 140 days of gestation and data have been collected from a total of 32 foetuses.

The histological preparation of the skin samples has been described previously (Claxton 1962).

(b) *Measurement of Spatial Pattern*

Clark and Evans (1954) described a measure of spatial relationships among individuals of any population, indicating the manner and degree to which their arrangement departs from a random one. They have defined a value, R , which varies according to $0 \leq R \leq 2.1491$, and which is given by the product of twice the square root of density and a mean of distances from each individual to its nearest neighbour. The departure of R from unity either towards the lower or upper bound indicates a departure of the distribution from a random one in the direction of aggregation or uniformity respectively. The description of spatial relationships among follicles is based on values of R calculated from follicle density and nearest neighbour measurements between follicles.

(c) *Measurements from the Follicle Population*

During histological preparation the skin material underwent shrinkage and various degrees of distortion. The follicle counts on which density estimates were based were adjusted for these changes with methods described by Carter and Dowling (1954).

Claxton (1962) compared two methods of adjusting interfollicle measurements for the effects of shrinkage and distortion, and found both gave similar results with equal accuracy. The more simple method of correction using a ratio of the original skin sample diameter to a mean of four equally spaced section diameters has been adopted here.

All recordings were taken as near as possible to the mid-sebaceous gland level from drawings of the follicle population made with the aid of a projection microscope. A mean distance to nearest neighbour for the primary follicles in each section was calculated from 10 distances using central primaries as centres and 16 distances using laterals as centres. These numbers are in proportion to the ratio $PL : PC$, which was found to be about 1.6 (Claxton 1962). The R values derived for the total follicle population are based on a mean distance to nearest neighbour using 40 individual distances, the follicle centres from which they were made being chosen from a random number series in a field containing about 100–140 follicles.

The R values which were subjected to analyses of variance were considered to be normally distributed. Several missing plots in the data were treated using methods given by Snedecor (1956).

III. RESULTS AND DISCUSSION

(a) *Accuracy of the Method*

Duplicate (adjacent) skin samples from each of five body positions on a foetus were used to estimate the errors in the values of R together with those in its components, density and mean distance to nearest neighbour. The duplicate material comprised that from one foetus of each breed at ages of 90 and 110 days. Because all measurements were recorded at mid-sebaceous gland level, the developing secondary follicle population was not visible in some of the 90-day material. Consequently the duplicates from the older foetuses only have been used for error estimates in the total follicle population.

TABLE I
ACCURACY WITH WHICH R , DENSITY ρ (AS FOLLICLES/MM²), AND MEAN DISTANCE TO NEAREST NEIGHBOUR \bar{r} (MM) HAVE BEEN MEASURED ON ANY POSITION OF A FOETUS

Age of Total Primary Population (days)	R		ρ		\bar{r}		Average No. of Follicles Counted per Section	No. of Distances Measured in Each Section
	S.D.*	C.V.†	S.D.	C.V.	S.D.	C.V.		
90	0.110	7.95	11.28	17.12	0.010	11.80	185	26
110	0.100	6.63	2.73	8.61	0.009	6.89	248	26
Total population	0.088	8.37	31.75	13.91	0.002	6.43	257	40

* S.D., standard deviation.

† C.V., coefficient of variation.

The standard deviations and coefficients of variation for the duplicate samples are shown in Table 1 and indicate the accuracy with which measurements have been made on any given position of a foetus. The mean number of follicles counted in each section to estimate density, together with the number of nearest neighbour measurements made in each section, is also given.

The high variation of density and of mean distance to nearest neighbour among primary follicles at 90 days has resulted mainly from marked differences in two pairs of duplicate recordings—the same pairs which were responsible for high error estimates in the measurements among central primary follicles (Claxton 1962). Moreover, in neither case were the errors in the resultant R values affected so that the two pairs are not considered typical. Their exclusion from the present series leaves coefficients of variation of 12.6 and 9.3% for density and mean distance to nearest neighbour respectively.

(b) *Density, Mean Distance to Nearest Neighbour, and R Values among Primary Follicles*

The mean values of R and their components derived from all primary follicles are summarized in Table 2. Measurements were made at six body positions on two foetuses of each breed at all ages except 70 and 90 days where one foetus and three foetuses of each breed respectively contributed to the results.

(i) *Values of R during Primary Initiation.*—The R values derived for all primary follicles are lower than those derived from central primaries alone, the mean of the differences [$R_{PC} - R_{(PC+PL)}$] being positive in both breeds at all ages. Thus the degree of uniformity in the distribution of primary follicles is lowered as their initiation proceeds due to the appearance of lateral primary follicles in the central primary population. A preliminary analysis of the variation (that between foetuses only) in the R values showed that differences between the breeds was not significant. Moreover, the R values at different ages were not dependent on the breed. Thus the spatial arrangement of primary follicles is similar and undergoes similar changes in Merino and Romney \times Southdown cross sheep during the period 70–140 days of foetal age.

TABLE 2
MEAN VALUES OF R , PRIMARY DENSITY ρ (AS FOLLICLES/MM²), AND MEAN DISTANCE TO NEAREST NEIGHBOUR \bar{r} (MM)

Each entry (except at 70 and 90 days)* a mean of six body positions on each of two foetuses

Age (days)	Romney \times Southdown			Merino			Mean
	R	ρ	\bar{r}	R	ρ	\bar{r}	R
70	1.60	44.4	0.122	1.66	43.9	0.126	1.63
80	1.39	50.6	0.100	1.41	64.9	0.089	1.40
90	1.46	56.7	0.099	1.43	72.5	0.086	1.45
100	1.49	38.0	0.124	1.52	53.1	0.106	1.50
110	1.51	30.3	0.139	1.53	32.2	0.137	1.52
120	1.48	20.7	0.166	1.53	25.7	0.154	1.51
130	1.48	17.6	0.183	1.56	21.0	0.175	1.52
140	1.50	13.2	0.212	1.54	16.6	0.191	1.52

* See text.

The age variation in R was significant ($P = 0.05$) and was due largely to low values of R at 80 days compared to higher values at 70 days. This suggests therefore that the values of R , rather than decreasing continuously as primary initiation continues, reach their lowest levels before initiation is complete. Whether or not these values rise again toward the end of initiation is not clear but an increase would be expected if two laterals did not appear (at a level in the skin where measurements were made)—simultaneously, on each side of the central primaries.

(ii) *Values of R following Primary Initiation.*—Because the relationship between stage of follicle initiation and foetal age varies over the body, the R values from 70- and 80-day-old material, before primary initiation was complete, were excluded from the complete variance analysis, which is shown in Table 3 for the remaining ages. The only significant F value ($P < 0.05$) is that associated with differences between body positions. The means of positions were subjected to Duncan's (1955) multiple-range test which showed that the back and neck regions yielded significantly higher R values than did the breech position.

At 90 days of foetal age at least, a greater proportion of nearest neighbours to the lateral primaries are primary follicles (almost always the central) belonging

to the same corresponding trio group. Variation in this proportion might also be expected to be associated with differences in the resulting R values. On six body positions from each of six 90-day-old foetuses, 40 lateral primary follicles, selected at random, were classified according to whether they were nearer to a primary member of the same, or of an adjacent, trio group. The percentages falling into the latter class are shown in Table 4 together with the analysis of variance on the transformed (arc sin) values. The differences between positions were found to be significant at the 1% level. Further, the transformed proportions on each position and the mean R values for these positions were found to be positively correlated ($r = +0.88$, d.f. = 4, $P < 0.05$).

TABLE 3
ANALYSIS OF VARIATION IN THE R VALUES DERIVED FROM PRIMARY FOLLICLES

Source of Variation	Degrees of Freedom	Mean Square	F
Between foetuses	25		
Age	5	0.0247	n.s.†
Breed	1	0.0316	n.s.
Age \times breed	5	0.1001	n.s.
Replication	14	0.0259	
Within foetuses	127		
Position	5	0.0148	2.361*
Position \times breed	25	0.011	n.s.
Position \times age	5	0.010	n.s.
Position \times age \times breed	25	0.006	n.s.
Error	67	0.006	

* $P = 0.05$.

† n.s., not significant.

Although differences in the distance relationships between primary follicles belonging to the same or to adjacent trio groups adequately account for the regional variation in R values, their cause has not been established. Because these differences appear so soon following initiation it seems more likely that they result during initiation rather than from changes occurring after initiation. This is also supported by the fact that the poll region which is the most advanced has not yielded extreme R values. Further, the back and midside positions, which differ markedly in the percentage of nearest neighbours belonging to adjacent groups, have been shown by Stephenson (1958) in New Zealand Romney and N -type sheep to commence follicle initiation at about the same time.

(iii) *Changes in Density and Mean Distance to Nearest Neighbour after Initiation.*—Because primary follicle initiation is complete at 90 days of foetal age (Schinckel 1955) later changes in primary density result from skin expansion (Carter and Hardy 1947; Stephenson 1958). These changes have been analysed by regressing the

logarithm of density on the logarithm of age for each position and in both breeds (cf. Stephenson 1958). A similar regression has also been carried out with the logarithm

TABLE 4
PERCENTAGES OF LATERAL PRIMARY FOLLICLES NEARER TO A MEMBER OF AN ADJACENT PRIMARY TRIO GROUP AT 90 DAYS, AND THE ANALYSIS OF THESE TRANSFORMED (ARC SIN) VALUES

Breed	Poll	Neck	Shoulder	Back	Midside	Britch
Romney \times Southdown	25	27	22	28	9	15
Merino	30	16	15	27	13	7

Analysis of Variance

Source of Variation	Degrees of Freedom	Mean Square	<i>F</i>
Between sheep	5		
Breed	1	13.3	
Error	4	269.0	
Within sheep	30		
Position	5	156.3	4.78**
Position \times breed	5	67.2	2.06†
Error	20	32.7	

** $P < 0.01$.

† Not significant.

TABLE 5
REGRESSION COEFFICIENTS FOR THE LOGARITHMS OF MEAN DISTANCE TO NEAREST NEIGHBOUR AND DENSITY AMONG PRIMARY FOLLICLES ON THE LOGARITHM OF AGE

	Poll	Neck	Shoulder	Back	Midside	Britch
Romney \times Southdown						
Density	-1.921	-3.587	-3.275	-3.706	-3.605	-3.315
Mean distance to nearest neighbour	0.821	1.793	1.699	1.931	2.022	1.820
Merino						
Density	-2.527	-3.809	-3.090	-4.067	-3.327	-3.292
Mean distance to nearest neighbour	1.259	2.120	1.715	2.188	1.917	1.814

of mean distance to nearest neighbour. The regression coefficients are shown in Table 5.

The coefficients derived from the regression of log density on log age and indicating the rates of skin expansion on different body positions are generally lower than those calculated from the changes in central primary density (Claxton 1962). However, they are considered to be more accurate as they were derived from measurements recorded in a greater number of foetuses covering a more comprehensive range of foetal ages. Skin expansion between 90 and 140 days of foetal age in both breeds was lowest on the poll, shoulder, and britch regions and highest on the midside, neck, and back regions. The rates of skin expansion in both breeds were generally less than those given by Stephenson (1958) for New Zealand Romney and *N*-type sheep. In comparison, the most notable feature of Stephenson's data was the higher rate of skin expansion on the poll and britch positions.

If, as the analysis of variance in Table 3 would suggest, the *R* values between 90 and 140 days are constant, the two variable components of *R*, root density and mean distance to nearest neighbour, would be inversely proportional to one another during this period. Thus the coefficients of regression for log mean distance to nearest neighbour on log age should be opposite in sign to, and half the value of, those derived for the corresponding regressions using density. Generally, however, the rates of skin expansion given by the decreasing densities on different positions were less than those derived from the increasing values of mean distance to nearest neighbour. Ignoring signs, the differences between the regression coefficients for density on each position in each breed and twice the corresponding values of those for mean distance to nearest neighbour were found to be significantly different from zero (Student's $t = 3.35$, d.f. = 11, $P < 0.01$). This implies therefore, that distances to the nearest neighbour of each follicle have increased beyond what might be expected to result from decreases in density, and that the positions of follicles have altered relative to one another.

Each nearest neighbour measurement which contributed to the *R* values was also scored according to whether the pair of follicles involved belonged to the same or to different trio groups of primaries. These results showed that the proportion of nearest neighbours which were members of the same trio group of primaries decreased with age from an average of 86% at 90 days, through 78 and 73% at 100 and 110 days, respectively, to 72% at 120 days.

Because observations were regularly made as near as possible to mid-sebaceous gland level, the measurements recorded in the older foetuses were further from the skin surface than those recorded in the younger foetuses. Moreover, it is commonly recognized that the follicles within each bundle, in adult sheep at least, noticeably diverge as they penetrate the dermis so that the identity of the follicle groups is usually lost in the region of the follicle bulbs. Thus there is a possibility that the changes in the relative positions of follicles observed at different ages could result from the comparison of follicle relationships at different absolute depths in the skin. This aspect was investigated in some of the 130-day-old material. From 12 sets of serial sections, representing a number of body positions in four different foetuses at this age, about 170 follicle groups were drawn at the characteristic mid-sebaceous gland level. Each group was subsequently traced upwards through the serial sections an average of about 200 μ and drawn a second time. The distances between

each central primary and its laterals were compared at the two levels and found to differ on average by less than 0.5% of the overall mean distance separating a central from a lateral. Thus any divergent growth between the skin surface and mid-sebaceous gland level, of follicles within the same group, appears to be negligible in foetal material.

The evidence indicates therefore that the rate of skin expansion within the area bounded by each group is greater than that between the trio groups. Also, because the proportion of lateral primary follicles which were nearest to a member of their own trio group did not drop below 50% (for $PL:PC = 1.63$, this would correspond to about 70% of nearest neighbours among all primary follicles being members of the same trio group), the R values between 90 and 120 days would be expected to increase. The mean values of R given in Table 2 are in fact suggestive of an upward trend after 90 days.

In several British breeds of sheep, Burns (1949, 1953) found that although occasional adjacent follicle groups became contiguous after birth, the associated increasing difficulty in identifying trio groups was more than offset by the deeper staining properties of collagen fibres between the groups which tended to define the follicle bundles more clearly. Carter (1940) noted that the grouping so characteristic among the follicles in foetal material was obscured in the adult, especially in the Merino where he suggested that crowding of the later generations of follicles rapidly distorted the original form of the grouping. In the present study the classification of primary follicles and the identification of primary trio groups became more difficult as the age of the foetus increased. It is suggested therefore that the differential movement between the primary follicles after their initiation contributes to this effect. The extent to which these changes continue after birth was not examined.

(c) *Density, Mean Distance to Nearest Neighbour, and R Values in the Total Follicle Population*

The mean values of R and their components, derived from the total follicle population, are given in Table 6. The same numbers of fetuses contributed to each age group as those previously indicated in the results for primary follicles. With the appearance of first secondary follicles at a level of the primary follicle sebaceous glands the value of R in the total follicle population dropped markedly and reached a level close to unity. The results show that the value initially reached in the Romney \times Southdown cross material is lower than the corresponding value in Merino fetuses. This difference at 100 days, however, was not significant and the spatial relationships between the follicles at this stage are considered similar in both breeds.

The distribution of nearest neighbour measurements from the total follicle population is bimodal. It appears to be a composite of two unimodal distributions—one arising from measurements using primary follicles as centres and the other from measurements using secondary follicles as centres. Therefore the R values derived from the total follicle population and from primary follicles alone may not be strictly comparable.

(i) *Values of R during Secondary Follicle Initiation.*—Because the earliest foetal age at which all sections showed some secondary follicle development at

TABLE 6
MEAN VALUES OF R , DENSITY ρ (AS FOLLICLES/MM²), AND MEAN DISTANCE TO NEAREST NEIGHBOUR \bar{r} (MM), IN THE TOTAL FOLLICLE POPULATION
Each entry (except at 70 and 90 days)* a mean of six body positions on each of two foetuses

Age (days)	Romney \times Southdown			Merino			Mean
	R	ρ	\bar{r}	R	ρ	\bar{r}	R
70	1.60	44.4	0.122	1.66	43.9	0.126	1.63
80	1.39	50.6	0.100	1.41	64.9	0.089	1.40
90	1.25	85.9	0.708	1.37	90.9	0.767	1.31
100	1.01	107.2	0.494	1.13	167.5	0.448	1.07
110	1.06	147.6	0.449	1.12	269.3	0.348	1.09
120	0.99	108.4	0.498	1.01	284.4	0.310	1.00
130	1.05	141.6	0.499	1.04	361.1	0.288	1.05
140	1.13	81.6	0.648	1.02	288.8	0.316	1.08

* See text.

mid-sebaceous gland level was 100 days, only the values of R at this and later ages were subjected to the variance analysis shown in Table 7. The F value of 3.83

TABLE 7
ANALYSIS OF VARIATION IN THE R VALUES DERIVED FROM ALL FOLLICLES

Source of Variation	Degrees of Freedom	Mean Square	F
Between foetuses	19		
Age	4	0.0267	n.s.†
Breed	1	0.0124	n.s.
Age \times breed	4	0.0449	3.828*
Replication	10	0.0117	
Within foetuses	97		
Position	5	0.0236	4.717**
Position \times breed	20	0.0054	n.s.
Position \times age	5	0.0104	n.s.
Position \times age \times breed	20	0.0045	n.s.
Error	47	0.0050	

* $P < 0.05$.

** $P < 0.01$.

† n.s., not significant.

corresponding to the interaction term between age and breed was significant at the 5% level. The highly significant ($P < 0.01$) differences between positions were

shown by the multiple-range test to result entirely from higher R values on the poll region. The smaller differences between the five remaining positions were not significant.

Any tendency for continued secondary initiation to alter the resultant values of R would be expected to depend upon the number of secondary follicles initiated in each primary trio group, i.e. on the ratio of secondary to primary follicles, S/P . The mean S/P ratios are shown in Table 8, each entry being the average of ratios in the corresponding position on two foetuses of the same breed and age. These values also suggest that the higher values of R on the poll region may result from the noticeably lower S/P values for the same region. Consequently an analysis of covariance allowing for the effects of S/P was performed on the R values between 100 and 140 days.

TABLE 8
VALUES OF S/P AT DIFFERENT AGES FOR SIX BODY POSITIONS IN BOTH BREEDS
Each entry a mean from two foetuses

Breed	Age (days)	Poll	Neck	Shoulder	Back	Midside	Britch	Mean
Romney \times Southdown	100	2.1	2.2	1.6	1.3	1.5	1.7	1.7
	110	2.3	4.5	4.0	3.4	4.0	3.6	3.6
	120	2.0	5.0	5.0	5.0	4.4	4.2	4.3
	130	1.9	6.2	6.7	6.3	8.6	5.1	5.8
	140	2.2	7.3	5.5	5.4	5.9	4.9	5.2
Merino	100	4.6	2.5	2.1	2.1	1.3	1.6	2.4
	110	8.6	6.1	7.7	6.7	6.5	6.1	6.9
	120	8.4	10.0	12.9	8.5	9.0	8.4	9.5
	130	8.1	14.5	17.0	19.4	18.8	14.6	15.4
	140	11.6	17.1	18.8	19.3	17.0	16.6	16.7

No term in the analysis of covariance was significant. The resulting reduction in the sums of squares for the interaction between age and breed was about 92% and the corresponding correlation ($r = 0.96$, d.f. = 3) was highly significant ($P < 0.01$). The sums of squares for positions was reduced by about 56% yet the correlation was not significant.

Although an association between R and S/P on different body positions has not been demonstrated, there is no evidence that R displays regional variation when adjusted for the ratio of secondary to primary follicles. Different trends in R in the two breeds between 100 and 140 days of foetal age are adequately explained by the different changes in S/P .

(ii) *Expansion of the Follicle Group.*—The first secondary follicles form between each lateral and central of the primary trio groups and subsequent initiation occurs largely between the early secondaries and the primaries (Carter 1943; Burns 1949; Ryder 1958). It was expected therefore that the continued initiation of secondaries

would result in a marked aggregating tendency which would be reflected in the value of R . This, however, has not been found and two possible reasons for this seem likely. Firstly, some secondary follicles are initiated on the outside edges of each lateral primary follicle. Secondly, the ability to recognize the first secondary follicles in later foetal life on the secondary margin of each follicle group, together with the fact that the secondaries later lie to one side of the primaries (Carter 1940; Ryder 1962), suggests that the secondaries move outwards from the primaries as initiation proceeds.

Table 9 shows ratios of the mean distance from each central primary to the first-formed secondaries and to the laterals of the same corresponding group. Each value is a mean of these ratios from seven follicle groups chosen at random within each section from those groups containing three primary follicles. Measurements were made in two fetuses of each breed at both 100 and 130 days of foetal age. An analysis of these ratios showed the difference at the two ages to be significantly ($P < 0.02$) different from zero.

TABLE 9
RATIOS WITHIN EACH FOLLICLE GROUP OF THE MEAN DISTANCE BETWEEN A CENTRAL PRIMARY AND THE FIRST-FORMED SECONDARIES TO THE MEAN DISTANCE BETWEEN THE SAME CENTRAL AND THE LATERAL PRIMARIES

Age (days)	Breed	Neck	Shoulder	Back	Midside	Britch
100	Romney \times Southdown	1.07	1.16	1.03	0.95	0.90
		1.04	1.20	1.28	0.93	1.15
	Merino	1.06	1.05	0.98	0.86	1.01
		0.91	0.98	0.94	0.94	1.02
130	Romney \times Southdown	1.47	1.22	1.34	1.43	1.55
		1.52	1.31	1.70	1.49	1.56
	Merino	1.07	1.02	1.26	1.38	1.20
		1.44	1.42	1.55	1.45	1.62

Therefore in addition to the differential movement of the laterals away from the central primary of each group there is a movement of the secondaries outwards from the primary follicles at an even greater rate. The resulting effect is that the relative area occupied by the groups of follicles increases as secondary initiation and development continues, and the tendency for the follicle pattern to become increasingly aggregated is largely counteracted.

It is not known whether the differential movement of secondaries results from a physical crowding effect or a localized growth stimulus. However, the observation that the relatively smaller and fewer secondary follicles in the skin of wild sheep tend to remain between the primaries (Ryder 1957, 1958, 1960) would favour the former alternative. Although the degree to which the early secondaries have moved outwards appears similar in the Merino and Romney \times Southdown cross material, the reduced crowding effect from fewer secondary follicles in the latter breed may be counterbalanced by their larger size.

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