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## The relationship between wool follicle density and fibre diameter is curvilinear

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### ABSTRACT

Records for wool follicle density and wool fibre diameter were obtained from ewes ( $n = 787$ ) of 13 breeds and lambs ( $n = 543$ ) of eight breeds and four crossbred combinations. The data were gathered as part of nine separate experiments from seven years. Mean fibre diameter ranged between 14 and 58  $\mu\text{m}$ , while follicle density covered the range 6.5 to 95 follicles/ $\text{mm}^2$ . There was a significant curvilinear relationship ( $P < 0.001$ ) between individual animal means for fibre diameter and follicle density. The relationship was significantly different between ewes and lambs ( $P < 0.001$ ). Furthermore, the relationship between breed means for density and diameter were compared with equivalent data from literature sources. The relationship was effectively the same for all sources of data ( $P = 0.455$ ). A restricted range of data could have led some literature sources to suggest a linear relationship!

**Keywords:** skin; sheep breeds; Merino; Romney; feral sheep

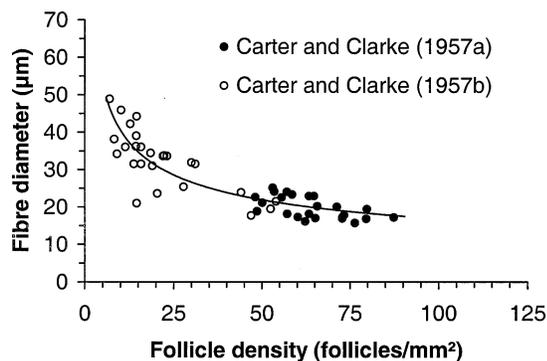
### INTRODUCTION

When analysing data by regression it is easy to be misled so that some underlying trend can be overlooked. This is particularly the case with the relationship between fibre diameter and follicle density, where follicle density is the total number of primary and secondary wool follicles in an area of skin. Working within a single breed, one can be distracted by the limits of the range of fibre diameter within the breed.

Carter and Clarke (1957a) published wool follicle density and fibre diameter data for a number of strains of Merino, and similar data for British breeds and Merino-based breeds in an accompanying paper (Carter and Clarke, 1957b). Fraser and Short (1960) took the data of Carter and Clarke (1957a,b) and published a figure to show the relationship between follicle density and fibre diameter. This data covered a wide range of diameters (16 to 49  $\mu\text{m}$ ) and follicle densities (7 to 87 follicles / $\text{mm}^2$ ). The relationship has been recreated in Figure 1 using the original data, with the Merino (Carter and Clarke, 1957a) distinguished from other breeds (Carter and Clarke, 1957b). From Figure 1 it is apparent that the Merino data alone could satisfactorily be explained by a straight-line relationship.

The following work was conducted to determine the form of this relationship in data collected in our laboratory, and to evaluate it in relation to published data.

**FIGURE 1.** The relationship between follicle density and fibre diameter from literature values for Merino sheep (Carter and Clarke, 1957a) and non-Merino sheep (Carter and Clarke, 1957b).



### MATERIALS AND METHODS

Records of wool follicle density and wool fibre diameter were obtained from ewes ( $n = 787$ ) of 13 breeds and lambs ( $n = 543$ ) of eight breeds and four crossbred combinations (see Table 1). The data were gathered as part of nine separate experiments that were carried out during seven years.

For lambs, the midside patch was cleared using a conventional shearing handpiece and a wool sample was then harvested within one month using Oster size 40 small-animal clippers. The lambs were slaughtered the following day and skin samples were obtained from the pelts after they were removed from the carcass at a commercial meat processor. The autopsy samples were collected using the biopsy trephine described by Scobie (1993), but the samples were taken from the flesh side of the skin, within the right midside region.

Scobie (1993) described the procedure for skin sampling from ewes. Wool samples were collected from right midside patches that were harvested each month for six months on the ewes. Skin biopsies were collected from the left midside. Although the left-side patch was harvested of wool each month, the wool samples were not suitable for measurement due to the biopsy from the previous month. The local animal ethics committee approved all procedures.

Skin samples were fixed and sectioned as described by Carter and Clarke (1957a). The number of follicles in 12 fields of 0.25  $\text{mm}^2$  were counted at sebaceous-gland level for each slide to estimate the mean number of follicles per square millimetre, or follicle density. The shrinkage of each section was estimated by measuring the area of the section on the microscope slide using image analysis (Video Pro 32, Leading Edge, Marion, South Australia), and adjusting the area of the section relative to area of the biopsy trephine.

Mean fibre diameter was measured (IWTO, 1998) using an OFDA (BSC Electronics, 1a Thurso Road, Myaree, Western Australia) on wool samples grown during the month prior to the collection of skin biopsy.

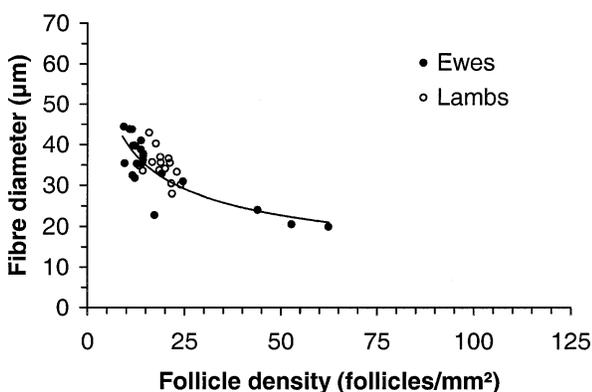
Alternative forms of the relationship between mean fibre diameter of wool grown during the month prior to biopsy sampling for follicle density, were determined using linear

and non-linear regression (GENSTAT 5, 1993). Statistical comparisons between alternative forms for the relationship were conducted by comparing the residual mean square for each.

### RESULTS

Mean fibre diameter ranged between 14µm and 58µm, while follicle density covered the range 6.5 to 95 follicles/mm+ (Table 1). There was a significant curvilinear relationship ( $P < 0.001$ ) between individual animal means for fibre diameter and follicle density. The relationship was significantly different between experiments depending on whether the animals were ewes or lambs ( $P < 0.001$ ).

**FIGURE 2.** The relationship between follicle density and fibre diameter for ewes and lambs of various breeds and crosses. The regression line for ewes ( $y = 92.2x^{-0.358}$ ) is shown.



Mean and standard deviation of fibre diameter and follicle density for each breed or crossbred, were calculated for each experiment. The relationship between mean follicle density and mean fibre diameter of these breeds is shown in Figure 2. The equation describing the relationship was  $y = 71.7x^{-0.261}$  (where  $x =$  density and  $y =$  diameter) for lambs ( $r^2 = 0.251$ ) and  $y = 92.2x^{-0.358}$  for ewes ( $r^2 = 0.539$ ). The standard errors of the estimates of the coefficients were 4.1 for lambs and 3.3 for ewes, and for the exponents were 0.019 and 0.014 respectively. A linear relationship  $y = 0.65x + 47$  accounted for more of the variation in lambs ( $r^2 = 0.443$ ), with standard errors of 0.02 and 1 for the coefficient and constant respectively.

**TABLE 1.** Follicle density and fibre diameter of various breeds of ewe and breeds and crosses of lamb.

Breed	Year	No. of sheep	Follicle density (Follicles/mm <sup>2</sup> )		Fibre diameter (µm)	
			Mean	SD	Mean	SD
<b>Ewes</b>						
Drysdale	1992	58	11.5	2.0	43.7	8.5
Drysdale	1993	25	9.4	1.4	44.4	6.7
English Leicester	1992	58	13.7	1.6	38.8	7.0
English Leicester	1993	10	10.8	1.9	43.8	3.2
Halfbred <sup>1</sup>	1992	60	24.6	3.6	30.9	4.9
Halfbred <sup>1</sup>	1993	24	19.2	3.0	32.9	2.8
Lincoln	1992	12	14.3	2.7	37.7	4.9
Lincoln	1993	10	13.8	3.4	41.0	3.9
Merino	1992	60	62.4	11.6	19.9	2.4
Merino	1993	24	52.8	9.0	20.5	1.4
Poll Dorset	1992	60	14.4	2.3	36.2	5.7
Poll Dorset	1993	25	12.1	1.5	39.8	3.0
Romney	1992	58	14.4	2.3	37.2	7.1
Romney	1993	55	12.3	2.6	39.7	3.9
Suffolk	1992	12	11.6	1.2	32.5	5.3
Suffolk	1993	10	9.6	2.0	35.4	3.9
Arapawa Island	1993	12	17.3	4.7	22.7	1.8
Cheviot	1993	45	13.9	2.1	35.1	3.3
Polwarth	1993	49	44.0	9.2	24.0	2.8
Shropshire	1993	50	12.7	2.1	35.3	3.5
Wiltshire	1993	69	12.2	2.4	31.8	2.7
<b>Lambs</b>						
Coopworth	1995 <sup>2</sup>	60	18.5	3.4	33.7	2.6
Drysdale	1994	10	11.7	2.5	39.7	2.7
English Leicester	1994	10	13.3	2.2	34.8	2.9
Halfbred <sup>1</sup>	1994	9	27.6	5.7	27.4	2.1
Lincoln	1995	11	15.9	2.9	42.9	1.9
Lincoln x Romney	1995	10	17.6	2.8	40.2	2.9
Perendale	1994	10	16.7	3.3	35.7	2.1
Romney	1994	9	14.1	0.8	35.5	1.4
Poll Dorset	1994	10	14.2	2.1	33.6	2.1
Poll Dorset	1995	12	21.3	2.6	35.5	2.4
Poll Dorset x Corriedale	1997	78	24.0	5.0	30.1	2.2
Poll Dorset x Romney	1995	27	20.9	3.3	36.6	2.4
Poll Dorset x Romney	1996	79	23.0	3.6	33.3	2.6
Poll Dorset x Romney	1997	40	21.8	3.7	28.0	2.5
Poll Dorset x Romney	1997 <sup>3</sup>	80	21.6	4.1	30.5	3.2
Poll Dorset x Romney	1998	38	20.0	3.2	34.1	2.3
Romney	1995	10	18.7	2.5	36.9	1.3
Suffolk x Romney	1998	40	18.9	3.6	35.6	2.1

<sup>1</sup> English Leicester x Merino interbred

<sup>2</sup> denotes a separate experiment from all others marked 1995

<sup>3</sup> denotes a separate experiment from all others marked 1997

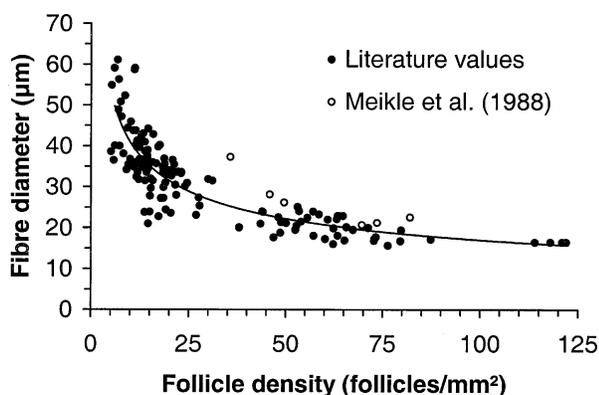
### DISCUSSION

Clearly, there was a curvilinear relationship between follicle density and fibre diameter for adult sheep. There was a significantly different relationship for individual animal data from lambs. The greater range of values for fibre diameter and follicle density in ewes has a strong effect on the shape of the curve in Figure 2, whereas there were no follicle density records above 25 follicles/mm<sup>2</sup> for lambs. In general, lambs tended to have a larger fibre diameter compared to ewes at an equivalent follicle density. It would be very interesting to extend the range for lambs to see whether they conform to a similar relationship to adult ewes, or to a straight line.

Figure 3 was compiled with data from our experiments, and numerous literature sources in which follicle density and fibre diameter were reported (Carter and Clarke 1957a; 1957b; Krishnarao *et al.*, 1960; Slee & Carter, 1962; Smith & Clarke, 1972; McGuirk *et al.*, 1978; Orwin & Whitaker, 1984; Steinhagen *et al.*, 1986; Bigham 1986; Massy, 1990;

Parry *et al.*, 1994; Pedraza *et al.*, 1994; Pearson *et al.*, 1999; Wuliji *et al.*, 1999). The pooled relationship for the ewes and lambs presented here ( $y = 89.8x^{-0.339}$ ) was not significantly different from the literature values ( $y = 98.6x^{-0.385}$ ) ( $P = 0.455$ ). The standard errors of the estimates of the coefficients were 12.2 for pooled data from lambs and ewes and 6.5 for the literature values, and for the exponents were 0.049 and 0.024 respectively. A large proportion of the variance (74 %) was accounted for by the general relationship  $y = 98.3x^{-0.380}$  (where  $x$  = density and  $y$  = diameter) for the results presented here pooled with those from the literature, with an overall standard error of 5.7 for the coefficient and 0.021 for the exponent. The results of Meikle *et al.* (1988) have been separately identified on Figure 3, and were excluded from the regression analysis since they seem to follow an entirely different curve. Meikle *et al.* (1988) made no mention of correction of the biopsy area for shrinkage (Carter and Clarke, 1957a; Steinhagen and Bredenhann, 1987), and the densities may be overestimated.

**FIGURE 3.** The relationship between follicle density and fibre diameter from literature sources (see text). The overall regression line ( $y = 98.3x^{-0.380}$ ) is shown.



Several important points arise from Figure 3. The data were collected in different laboratories from different breeds or strains, from animals of different ages bred in varied environments, yet they largely fit the same trend. Furthermore, those data points some distance from the curve fall into the low density, low diameter part of the plot rather than the high density, high diameter part. The breeds that relate to these points are the feral sheep from Orwin and Whitaker (1984), Pearson *et al.* (1999) and from our laboratory. Others are the Nilgiri (13.7, 23.8 µm) (Krishnarao *et al.*, 1960), the Suffolk (20.4, 23.6 µm) (Carter and Clarke, 1957b) and the Cheviot (14.6, 21.0 µm) (Carter and Clarke, 1957b). Other data points closer to the curve also represent the latter two breeds. The Cheviot from Steinhagen *et al.* (1986) (12.2, 36.2 µm) and from the data presented here (Table 1) had a mean fibre diameter greater than 21 µm. Similarly the Suffolk has been observed with a larger fibre diameter in our laboratories (Table 1). The breeds in this part of the plot also tend to be those with low fleece weight, and perhaps selection for fleece weight has increased density on breeds like the Romney which have a similar diameter but higher density. Conversely, natural selection may have reduced the density on the feral breeds,

so they produce lighter fleeces to survive unshorn in the feral state.

Woolliams and Wiener (1980) presented a figure similar to Figure 3 and described a curve of the same form  $yx^c = k$  (or  $y = kx^{-c}$ ) where  $x$  = density and  $y$  = diameter. They did not present the value for  $k$ , but the  $c$  value was very different to that of Figure 3 (2.31). Approximating the data from the information Woolliams and Wiener (1980) provided, the points lie well within the range presented in Figure 3. Their data were limited to densities of 14 to 50 follicles/mm<sup>2</sup> and this may have precipitated the difference in size of the exponent.

A restricted range of diameter could also have led Moore *et al.* (1989) to conclude that "... a highly significant negative and linear correlation ( $r = -0.89$ ) was found to exist between these two parameters." If we accept this, and extrapolate from their data (their Figure 5), then at around 37 µm fibre diameter, follicle density would be around zero follicles/mm<sup>2</sup>. Clearly this is untenable as a large portion of the New Zealand wool clip would not exist.

Many single trait selection experiments that might have indirectly increased fleece weight have been thwarted by the fact that other traits counteract the potential increase (reviewed by Davis and McGuirk, 1987). In pursuit of higher fleece weight and finer fibres, we may have ignored an underlying biological limit to fibre production that is outlined below.

The work of Galpin (1948) may help understand the relationship between follicle density and fibre diameter. Galpin (1948) found that the area of skin surface occupied by fibre was 2 square millimetres of fibre per square centimetre of skin for ewes. In other words, only two percent of the total available area was fibre cross section. Using follicle density and mean fibre diameter, this proportion was 1.7% for all the values presented in Figure 3. For the data from Figure 2 the value was 1.5% for ewes and 1.8% for lambs. Pooling 38 separate breed mean values for Merino sheep from Table 1 and literature, 2% of the surface of the skin was occupied by fibre on average. The average for Romneys was 1.6% calculated from five breed means (Table 1 and Bigham (1986)). Of course the calculation based on the square of mean fibre diameter will underestimate mean cross-sectional area of the fibres, where the distribution of fibre diameter is not normal but is skewed with a long tail in the high fibre diameter part of the distribution (Gilmour and Atkins, 1992). Nevertheless, there may be some biological ceiling on the area of skin that can be occupied by wool follicles. This hypothesis helps explain the good fit of the hyperbolic curve. The number of follicles per unit area of skin can vary from numerous and small to few but large, provided no more than about two percent of the surface is occupied.

Figure 3 presents a dilemma for sheep breeding. If the relationship is asymptotic, producing finer and finer fibres may be a challenge. To double density from 25 to 50 follicles/mm<sup>2</sup>, reduces diameter from 29 to 22 µm. Doubling density again will only reduce diameter to around 17 µm. The quest to produce the "Ultrafine Merino" has developed a very high density of around 120 follicles/mm<sup>2</sup> (Wuliji *et al.*, 1999), but extrapolating from the curve in Figure 3, about 200 follicles/mm<sup>2</sup> will be needed to produce

13  $\mu\text{m}$  fibres. Some individual sheep produce fibres of this mean diameter and it would be most interesting to check the actual follicle density.

The part of the curve for which there is no data, between 30 and 40 follicles/ $\text{mm}^2$  and between 20 and 30  $\mu\text{m}$  in diameter, is also intriguing. In the absence of data in these critical regions, the relationship could be composed of two straight lines through separate populations for the Merino and the "other" breeds. This is unlikely, since numerous individual ewes and lambs had values that lay in this region and our historical tendency to label sheep with a breed may be less statistically sound than pooling them into density or diameter classes.

Unusual genotypes may challenge this relationship. Unfortunately Short (1958) gave only the range of diameter (18 to 22  $\mu\text{m}$ ) and density (25 to 45 follicles/ $\text{mm}^2$ ) for "lustre" mutants, but the mean for this "breed" must lie in an unoccupied region of the plot. We were fortunate to obtain wool and skin from a "woolless" Suffolk sheep (Fiona Buchanan personal communication) that produced exceptionally fine wool for this breed (19  $\mu\text{m}$ ) from 13 follicles/ $\text{mm}^2$ . This Suffolk produced very little fleece (600g) relative to its live weight (100 kg). The fact that the fleece weight of both these types of mutants was much reduced makes them similar to the feral breeds discussed above. It may be both scientifically fulfilling and commercially valuable to find other animals or develop strains that also fall outside the constraints of this relationship.

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