

# THE EFFECT OF NUTRITIONAL DEPRIVATION IN EARLY FETAL LIFE ON FOLLICLE CHARACTERISTICS IN THE MERINO LAMB

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## SUMMARY

This experiment was conducted to test whether nutritional deprivation of a ewe during early gestation would affect the density of the initiating primary follicles and hence influence secondary follicle numbers and ultimately fibre characteristics of the mature animal. Pregnant ewes were denied access to feed and water for 96 hours at either 27 days or 35 days of pregnancy, or at both times. The lambs born from these ewes were measured for primary follicle density, secondary to primary follicle (S/P) ratio, and follicle density at 7 months of age. Wool growth and fibre diameter were measured at 7, 13 and 19 months of age. No significant differences were induced in any of the follicle traits. Lambs resulting from the ewes starved in only the late treatment period had higher primary follicle density (4.20 vs 3.77 follicles/mm<sup>2</sup>), lower S/P ratio (14.92 vs 17.42) and lower follicle density (6.173 vs 68.14 follicles/mm<sup>2</sup>) although these differences were not significant ( $P > 0.05$ ). Wool growth per unit area was significantly higher ( $P = 0.01$ ) in this group (8.29 vs 6.65 g/m<sup>2</sup>.day). Although not conclusive these results support the hypothesis that primary follicle density cannot be increased environmentally without a decrease in S/P ratio.

**Keywords:** primary follicle density, wool, sheep.

## INTRODUCTION

A major problem facing breeders of wool sheep is how to decrease average fibre diameter while maintaining or improving clean fleece weight. Progress is slow when selecting for both of these characteristics simultaneously, because there is a moderately high positive genetic correlation (about 0.3) between average fibre diameter and clean fleece weight (Mortimer 1987). Interest has focused recently on the biology and arrangement of wool follicles in the skin of the sheep because there may be a physiological basis for the genetic correlation between fleece weight and fibre diameter.

Under selection, increases in fleece weight have resulted from increases in follicle density (N), average fibre diameter, and fibre length rather than increases in wool growing surface area (Dun 1958; Turner 1958; Turner *et al.* 1968). Selection on any 1 of the components of wool weight causes compensatory changes in the others, resulting in little change in wool growth per unit area (Davis and McGuirk 1987). For example, selecting for and against N alone resulted in a 45% difference in N but very small difference (6.9%) in fleece weight (Turner *et al.* 1970).

If we can understand the physiological basis for the correlation between fleece weight and fibre diameter it may be possible to suggest additional selection criteria which will allow more rapid changes to be made in both traits.

Breeding lines were established to test if the effect of increasing N, by increasing primary follicle density (N<sub>p</sub>), was the same as increasing N by increasing secondary to primary follicle (S/P) ratio (Rendel and Nay 1978). From the responses in these selection lines Rendel and Nay (1978) concluded that high N resulting from high S/P did not result in increases in wool weight. They were unable to determine the effect on wool weight of increasing N by increasing N<sub>p</sub> as "changes in N<sub>p</sub> were accompanied by changes in the opposite direction in S/P ratio" (Rendel and Nay 1978). Thus, high N<sub>p</sub> and low N<sub>p</sub> lines had the same overall N. As a result of their studies Rendel and Nay (1978) proposed that selecting simultaneously for both primary follicle density and the ratio of secondary- to primary follicles may increase follicle density in a way that also increases clean fleece weight.

To study the physiological basis for the correlation between average fibre diameter and fleece weights, we have mimicked the predicted genetic changes using environmental means. It is possible to inhibit secondary follicle initiation and development on a lamb by severe under nutrition during pregnancy (Corbett 1979). There is less certainty about whether N<sub>p</sub> can be influenced in the same way. When low N<sub>p</sub> has been induced environmentally, any effect N<sub>p</sub> has had on wool growth has been confounded by low birth weight and poor ewe condition (Schinckel and Short 1961; Taplin and Everitt 1964). In these past experiments S/P ratio has also been directly affected by the nutritional treatment. However as wool follicles form in waves, primaries forming first, and secondaries forming relatively late in fetal life

(Fraser and Short 1960), it should be possible to isolate the effect of nutritional deprivation on primary follicle density.

This experiment was designed to test whether a short period of starvation prior to the time of primary follicle initiation will reduce Np without any subsequent effect on the S/P ratio. If successful this effect will allow us to test whether increasing N, by increasing Np and holding S/P constant, will result in increased fleece weight.

MATERIALS AND METHODS

One hundred and eighty mixed strain, medium wool ewes were divided into groups of 30 which were individually mated in paddocks to 6 different sires in late September 1991. Thirty six ewes, 4 tooth and older, were selected from this group based on time of joining using presence of raddle marks in a 48 hour period. The ewes had an average condition score of 4 and were increasing in liveweight. Seventeen days after joining the selected ewes were moved to individual pens in the animal house at the University of Western Australia for acclimatisation.

Table 1. Treatment means (adjusted for sire, sex, and litter size) of primary follicle density (follicles/mm<sup>2</sup> at 7 months of age), secondary to primary (S/P) follicle ratio (at 7 months of age), follicle density (follicles/mm<sup>2</sup> at 7 months of age), wool growth per animal (g/day measured up to 19 months), average fibre diameter (µm as measured at the 19 months shearing) and wool growth per unit area (g/m<sup>2</sup>.day grown between 13 and 19 months of age)

	Treatment group			
	Control	Starved period 1 only (starved early)	Starved period 2 only (starved late)	Starved both periods (starved both)
Number of lambs	5	4	8	6
Primary follicle density	3.80 (0.36) <sup>A</sup>	3.70 (0.42)	4.20 (0.36)	3.80 (0.34)
S/P ratio	17.06 (1.34)	17.83 (1.55)	14.92 (1.32)	17.36 (1.25)
Follicle density	67.6 (5.00)	68.39 (5.78)	61.76 (4.94)	68.42 ( 4.65)
Wool growth per head	6.21 (0.43)	6.38 (0.53)	7.16 (0.38)	7.06 (0.36)
Average fibre diameter	19.62 (0.78)	19.88 (0.96)	20.82 (0.69)	20.64 (0.66)
Wool growth per unit area	6.16 (0.41)	7.11 (0.49)	8.29** (0.35)	6.98 (0.37)
** Significantly different within this row (P = 0.01).				
<sup>A</sup> Standard error of the mean.				

A short period of starvation was applied to the pregnant ewes before the initiation of the first wave of primary wool follicles on the fetus. Two time periods were tested. In the early starvation period (period 1) 9 ewes were denied access to feed and water for 96 hours from days 27 to 31 of pregnancy, and in the late starvation period (period 2) 9 ewes were denied access to feed and water for 96 hours from days 35 to 39 of pregnancy. Nine control ewes were not starved in either period and a group of 9 animals were

starved in both treatment periods. Following the treatment period animals were fed 500 g of a 10% lupins and chaff diet for 1 day and 750g of this diet for a further 3 days. Following this ewes were returned to the paddock and run together.

Ewes were lambled indoors and following lambing, ewes and lambs were run outdoors on unimproved irrigated pastures with lupin supplementation until weaning (at 3 months of age). Thereafter, lambs were run in paddock conditions.

Lambs were shorn at 7, 13 and 19 months of age. Average fibre diameter, yield and fibre length were measured from midside samples taken at these shearings, and clean and greasy fleece weights were recorded. The tattoo patch method (Langlands and Wheeler 1968) was used to determine wool growth per unit area between the 7 and 13 months shearings and between the 13 and 19 months shearings.

Repeated measures analysis of variance was carried out on the wool traits. The model used included sire, sex, litter size and treatment group.

Skin samples (1 cm diameter) were taken at 7 months of age, fixed in paraffin wax and stained with haematoxylin, eosin and picric acid as described by Maddocks and Jackson (1988). Horizontal skin sections (7  $\mu\text{m}$ ) were taken and analysed for follicle density and primary follicle density at the sebaceous gland level. Follicles not producing fibres were also counted. Corrections were made for shrinkage on each individual sample, and the secondary to primary wool follicle ratio was calculated from the 2 density measurements. On average 130 primary follicles and 2000 secondary follicles were counted per animal.

## RESULTS

All 46 ewes in the experiment gave birth to live lambs. Of these, 26 lambs survived to the 7 months shearing. Lamb deaths were due to mis-mothering (50%), post-weaning losses (35%) and unknown causes (15%). Lamb deaths were not related to treatment group. There was no significant difference in the mean birthweight of the 4 groups (average 4.2 kg). The liveweights of the 4 groups were not different at 7, 13 and 19 months of age.

There were no significant differences between the control and treated groups in any of the follicle characteristics measured (Table 1). However, several trends consistent with expectations were observed. Lambs which were subjected to the late starvation period only (period 2 only), had the highest primary follicle density. This group also had the lowest S/P ratio and the lowest overall follicle density.

Clean fleece weights and average fibre diameters were not significantly different across groups, although lambs in groups which were starved at the latter time period (ie. period 2 only and both periods) grew more wool per day (7.11 vs 6.30 g/day) and had larger average fibre diameters (20.73  $\mu\text{m}$  vs 19.75  $\mu\text{m}$ ) in comparison to animals not starved in the latter time period. Lambs subjected to the late starvation period only had significantly greater ( $P = 0.01$ ) wool growth per unit skin area than other animals in the experiment (8.29  $\text{g/m}^2\cdot\text{day}$  vs 6.65  $\text{g/m}^2\cdot\text{day}$ ).

## DISCUSSION

### *Effectiveness of treatment*

Starvation in period 1 (early starvation) appeared to have no effect on primary follicle density or any other of the other characteristics measured in this experiment. This would suggest that primary follicle initiation cannot be affected by a short period of under nutrition prior to 35 days of fetal life. We may speculate that the mechanism determining the spacing of primary follicles is not active before this time.

Although not significant, the primary follicle density was higher in the group given only the late starvation. While the lack of significance is possibly because of the low numbers in the experiment, the increase in primary follicle density with under nutrition in early fetal life is consistent with the observations of Schinckel and Short (1961).

It is possible that when ewes were starved for both periods the late period of starvation had little effect, because of chance effects or because the longer period of starvation caused the ewe to mobilise body reserves, effectively buffering the fetus against nutritional deprivation.

### *Implications of lowering primary follicle density on other follicle characteristics*

The late starvation only group had the lowest S/P ratio and lowest follicle density of all the groups. If we assume that this treatment had no effect on the formation of secondary follicles, the data suggest that there is a physiological relationship between primary follicle density and the number of secondary follicles produced on a fetus. That is, as primary follicles are spaced closer together there are less secondaries formed in the space between them. The extension of this argument is that an increase in primary follicle density cannot be induced environmentally without a corresponding decrease in the S/P ratio. Adelson (1991) has shown that decreasing primary follicle density results in an increase in S/P ratio.

### *Fibre implications*

Animals in the late starvation only group had increased wool growth per unit area. As with Rendel and Nay's (1978) experiment there were corresponding decreases in S/P ratio and follicle density with increases in primary follicle density. Changes in wool growth cannot be attributed solely to changes in S/P ratio, follicle density or primary follicle density as all of these factors changed to some extent.

### *Existing models*

The above results may be explained by the reaction diffusion mechanism as presented by Nagorcka and Mooney (1985). In terms of the reaction diffusion model the observed increase in primary follicle density due to the late starvation treatment would be explained by a decrease in the initial spatial wavelength determining the distance between the first initiating primary follicles.

At this stage calculations of the rate of decrease in wavelength with time have not been estimated from this data set. However, if the rate of decrease of the spatial wavelength with time is related to the mitotic activity of the basal epidermal cell layer (Nagorcka and Mooney 1985), then we would expect it to be constant between the treatment groups in this experiment. If this is so, then we would expect a lower S/P ratio to occur with higher primary follicle densities, as was the case in this experiment.

In terms of the reaction diffusion model, for animals to be selected for both higher S/P ratio and higher primary follicle density, one possibility is that they would require decreases in the initial spatial wavelength and also increases in the rate of decline of the spatial wavelength. We may speculate, therefore, that the basis of the correlation between primary follicle density and S/P ratio is because the rate of decrease of the spatial wavelengths is not closely related to the initial spatial wavelength.

This experiment, while not conclusive, provides a further insight into the biological mechanism underlying the phenotypic association between clean fleece weight and average fibre diameter.

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