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The Effect of Supplements of Different Forms of Sulphur to the Diet upon the Wool of Merino Sheep.

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INTRODUCTION.

Considerable prominence has lately been given to the effect of adding sulphur supplements to the diet of sheep. Whatever the results may be from a nutritional or physiological point of view, the possible effect on the wool, as regards both quality and total production, naturally assumes importance, and should be investigated. This aspect was first remarked upon by Steyn (1931, 1932), who concluded that a sulphur supplement in the diet had a beneficial effect on the total production of wool. Subsequently Seddon (1933) recorded that he had found no significant effect on the wool yield of sheep, entirely grazed, while Pierce (1933) found no beneficial effect on sheep kept on a maintenance ration, or one slightly above maintenance.

The following are the results of an investigation of the wool of sixty Merino wethers which had received supplements of sulphur in various forms. The influence on the body weights of the same sheep has been dealt with in another publication (du Toit, Malan, Groenewald and Botha, 1935), where it was shown that, under the conditions of the experiment, the supplements had no significant effect on the body weight as compared with the control sheep. For this and other aspects of the question, it is only necessary to refer to the above publication, where full details will be found.

EXPERIMENTAL DETAILS.

A. Sheep and Management.

A group of seventy-five Merino wethers, born at Grootfontein between 10.3.1930 and 9.6.1930, were selected for uniformity of type, fleece, and body weight. They were shorn between 31.3.1931 and 8.4.1931, and subsequently sent to Onderstepoort, where they arrived on 8.10.1931 as two-tooths. Here they were shorn in December, 1931, and sixty of them placed on a basal ration for eleven months. During this (pre-experimental) period all the sheep received identical treatment, but were divided into six uniform groups and placed in a gravelled paddock with pens and boxes for individual feeding. At the conclusion of this period (November, 1932), they were shorn again and placed on the same basal ration for a period of twelve months, during which the sheep in five of the groups were dosed with various forms of sulphur, the sixth group acting as a control. They were finally shorn in November, 1933. For brevity the two periods will be called the 1932 and 1933 periods respectively. The conditions under which the wool was grown may thus be summarised as follows:—

1932: Eleven months on basal ration.

1933: Twelve months on basal ration and supplements.

B. Feed.

The basal ration throughout the two periods 1932 and 1933 consisted of half a pound of crushed yellow maize and half a pound of green feed—lucerne or maize stalks or maize ensilage during about three months in winter, when no green feed was available. In addition the animals had free and continual access to teff hay. For further details of the method of feeding and the consumption, as also of the general care and management of the sheep, the reader is referred to the paper quoted above (du Toit *et alia*, 1935).

During the experimental period (1933) the sheep were dosed daily except Sundays, the following being the supplements given to each animal:—

Group I: No supplement.

Group II: 0·45 gm. cystine.

Group III: 0·7 gm. of a mixture consisting of 0·2 gm. K_2SO_4, 0·3 gm. $MgSO_4$, and 0·2 gm. $CaSO_4$.

Group IV: 0·36 gm. KCNS.

Group V: 5·0 gm. sulphur.

Group VI: 0·12 gm. sulphur.

The quantities of supplements, except in the case of Group V, were calculated to contain the same amount of sulphur as that contained in the 0·45 gm. cystine given to each animal in Group II.

C. Material.

Immediately before shearing in December, 1931, a wool sample of about 15 gm. was cut from the shoulder of each sheep. A pair of fine scissors was employed, and the wool cut as close to the skin as possible. The area of about one and a half inches square, from which the sample had been cut, was then tattooed on the skin, and at subsequent shearings the wool growing on this area was first removed. In addition a further sample was taken adjacent to this area for chemical analysis, which is still in progress. This method ensured that the samples obtained at the end of successive periods were taken from the same area on the skin of each sheep.

The samples having been removed, the sheep were shorn and the fleeces stored in linen bags for examination.

D. Methods of Analysis.

(*a*) The grease weight, clean weight and yield percentage of the fleece.

At the time of shearing each fleece was properly skirted in the manner customary in shearing sheds. The skirtings were transferred to a smooth-topped table and again worked through carefully—heavy dung-locks were discarded, while all the lightest pieces that would blend with the fleece were run in with the latter. Fleeces and locks, individually, were then transferred to suitable linen bags and stored in a non-ventilated room. Subsequently these were weighed under identical conditions, due allowance being made for the weights of the shoulder samples which had been removed for physical and chemical analysis.

For the determination of the clean weights and yield percentages of individual fleeces, a modification (Botha) of the method described by Wilson (1928) was followed. The method may be summarised as follows:—

(1) Subsequent to the weighing referred to above, each fleece was passed three times through a fleece-breaking machine, the object being to blend the various parts of the fleece thoroughly in order to enable representative samples to be drawn from it.

(2) The fleece was weighed again to allow for impurities lost during the process of blending and for probable fluctuations in moisture content.

(3) Immediately after this weighing two samples of a hundred grams each were made up of small amounts of wool, of approximately twenty grams, taken at random from various parts of the bulk of the fleece.

(4) These samples were next scoured in a scouring liquor consisting of a mixture of potassium oleate soap and sodium carbonate.

(5) After being allowed to become air-dry, the air-dry weights of the samples, and of a standard sample of known dry weight—in hygroscopic equilibrium—were recorded.

(6) Immediately after this a sub-sample of twenty grams of air-dry wool was taken from the last-scoured sample of each fleece and dissolved in 200 c.c. N/NaOH solution (i.e. 4 per cent.), in order to determine the amount of vegetable matter present after scouring. The clean dry weights of the fleeces could then be calculated and converted to clean conditioned weight at 17 per cent. regain (International regain). The clean conditioned weight of wool contained in the locks was obtained separately in a slightly modified manner.

Next the air-dry grease weight of the locks was added to that of the corresponding fleece, and similarly the clean conditioned weight. From the resulting totals the weighted mean of the conditioned yield percentage of each total fleece was calculated.

The grease and clean conditioned weights given below represent the mean production over twenty-four hours.

The conditioned yield percentage is defined as the conditioned clean weight of the fleece expressed as a percentage of the original air-dry grease weight of fleece prior to machine treatment.

NOTE.—All grease weights have been expressed on an air-dry basis irrespective of actual moisture content. A higher or lower moisture content of the grease wool at the time of the initial weighing would be reflected merely in a lower or higher yield percentage. Such differences in the actual moisture content of the greasy wool could be brought about (*a*) by differences in atmospheric temperature and humidity in the different years at the time of weighing individual fleeces and (*b*) by the possibility that when the fleeces were stored in a closed non-ventilated room, equilibrium of moisture content may not have been attained.

It must be emphasised, however, that, by the nature of the calculations, the result for the conditioned clean weight remains totally unaffected by fluctuations of the moisture content of the greasy wool, no matter how wide these may have been. Even in the case of yield percentages, differences will be of the order of a few per cent. only, since the actual fluctuations in humidity will be comparatively small.

(*b*) Mean length, thickness and fibre weight of a sample.

The weight-length method (Roberts, 1930), was used for the analysis of the samples. It consists briefly of (1) a method of sampling, (2) the counting of a number of fibres, (3) the measurement of the lengths of a certain proportion, and (4) the determination of the dry weight of the fibres counted. From the data thus obtained the following are calculated:—

Mean length : l,

Mean thickness : $\sqrt{\dfrac{4W}{\pi \times 1\cdot3 \times n \times l}}$.

Mean fibre weight : $\dfrac{W}{n}$,

where W = dry weight of the counted fibres,

n = number of fibres counted,

l = mean length of one-tenth of the counted fibres,

$1\cdot3$ = specific gravity of dry wool.

In addition, a check on the sampling and on the accuracy of the result is obtained in the form of a coefficient of fineness dispersion. It should be pointed out that Roberts expresses his results as fibre fineness, which is the length, in centimetres, of fibre which weighs one milligram. The thickness given in this paper is the diameter of the fibres calculated on the assumption that they are uniform circular cylinders.

The samples were divided into ten approximately equal zones, and ten sub-samples made up, each consisting of one small lock from each zone. These sub-samples therefore collectively consisted of a hundred small locks taken from different points in such a way as to represent the whole of the marked area. Each sub-sample was analysed separately, so that ten different estimates of the length, fineness and fibre weight of the original sample were obtained. About two thousand fibres in all were counted from each of the pre-experimental samples, and four thousand from each of the experimental samples, the coefficients of fineness dispersion ranging from one to four per cent. The length of every tenth fibre was measured, and the dry weights were determined by comparison with a standard sample of known dry weight, the weighings being repeated daily for several days in succession.

The microscopical method of measuring thickness (Deurden, 1929), was applied to a number of the samples. This method gave essentially the same results, though the values were all somewhat higher; when routine analysis have to be made the authors are inclined to favour the microscopical method, especially in the case of Merino wool, where the fibres are very fine. A further comparison of the two methods, involving measurements on about a hundred samples, will be reserved for a later publication.

RESULTS AND STATISTICAL ANALYSIS.

Below are given the group means for the two periods, 1932 and 1933. A method of analysis of variance and co-variance (Fisher, 1932), has been employed to test the significance of group differences. It is realised that in connection with wool a very large proportion of the variation between different sheep under similar conditions is due to what may be described in general terms as individuality, and is independent of normal nutritional treatment. This part of the variance of values of the experimental period may be regarded as having been foreshadowed in the values of the pre-experimental period. The appropriate coefficient of linear regression is given by the ratio of the co-variance to the variance of the independent variate, which in this case is the variance in the preliminary values.

(*a*) Grease weights of the fleeces, in gm. calculated for one day's growth.

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932...............	10·7	10·7	9·6	10·8	9·9	10·8	10·4
1933...............	14·2	14·3	12·9	14·0	13·7	14·0	13·8

The sums of squares and products are given in the following table.

x_2 = Grease weight in 1932.
x_3 = Grease weight in 1933.

	Degrees of freedom.	x_2^2	x_2x_3	x_3^2
Between groups................	5	12·51	10·25	10·92
Within groups..................	47	73·43	76·55	124·03
TOTAL...............	52*	85·94	86·80	134·95

* There was one value missing in each of the two years. These were supplied by calculating the best-fitting value from the data for three years (Yates, 1933). This accounts for the fact that these is one degree of freedom less.

The result of applying the correction for the regression, as explained above, and analysing the variance of the adjusted yields, may be derived directly from the analysis of sums and products already presented. If b stands for the regression coefficient, comparisons of the adjusted values will be comparisons of quantities $(x_3 - bx_2)$. Now

$$(x_3 - bx_2)^2 = (b^2x_2^2 - 2bx_2x_3 + x_3^2).$$

In the present example,

$$b = 1\cdot0425$$
$$-2b = -2\cdot0850$$
$$b_2 = 1\cdot0868$$

and applying this directly to the sums of squares and products above, the following analysis is obtained.

Analysis of adjusted grease weights :—

	Degrees of freedom.	Sums of squares.	Mean squares.
Between groups................	5	3·1446	0·6289 = V_1
Within groups..................	46*	40·973	0·8907 = V_2
TOTAL...............	51	44·1176	—

The ratio $R = V_1/V_2$ is used to find the significance of the difference between the two variances (Snedecor, 1934). In the present case,

$$R^{-1} = \frac{8907}{6289} = 1 \cdot 416,$$

$n_1 = 46,$

$n_2 = 5,$

which is very insignificant and does not indicate any differential effect between the treatments.

(*b*) Clean weights of the fleeces in grams, calculated for one day's growth.

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932................	4·76	4·86	4·53	4·94	4·60	5·23	4·82
1933................	6·47	6·53	6·26	6·59	6·55	6·86	6·55

The same procedure has been followed in analysing the clean weights, with the following results:—

y_2 = clean weight for 1932.

y_3 = clean weight for 1933.

Sums of Squares and Products.

	Degrees of freedom.	y_2^2	y_2y_3	y_3^2
Between groups................	5	2·8718	1·9453	1·6702
Within groups..................	47	17·8063	16·8198	30·2822
TOTAL...............	52	20·6781	18·7651	31·9525

$b = 0 \cdot 9446.$

Analysis of Adjusted Values.

Variance.	Degrees of freedom.	Sum of Squares.	Mean Squares.
Between groups................	5	0·5576	0·1115
Within groups..................	46	14·3942	0·3129
TOTAL.................	51	14·9518	—

$R^{-1} = 2 \cdot 806.$

$n_1 = 46.$

$n_2 = 5.$

There is again no significant difference between the variances and consequently there is no reason for assuming that the clean weights were affected by the sulphur supplements.

(*c*) Yield per cent. of the fleeces:—

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932................	44·5	45·7	47·4	45·9	46·5	48·1	46·3
1933................	45·5	45·6	48·7	47·3	48·0	49·2	47·4

From the above analysis it is clear that no significant value as regards yield per cent. can be expected.

z_2 = yield per cent. for 1932.

z_3 = yield per cent. for 1933.

Sums of Squares and Products.

	Degrees of freedom.	z_2^2	z_2z_3	z_3^2
Between groups................	5	77·52	85·23	107·31
Within groups..................	47	841·27	555·30	601·50
TOTAL...............	52	918·79	640·53	708·81

$b = 0{\cdot}6601.$

Analysis of Variance of Adjusted Values.

Variance.	Degrees of freedom.	Sums of squares.	Mean squares.
Between groups................	5	28·5733	5·5147
Within groups..................	46	234·946	5·1075
TOTAL...............	51	263·5193	—

$R = 1{\cdot}0797.$
$n_1 = 5.$
$n_2 = 46.$

As was expected, the variance between groups is not significantly different from that within groups.

(d) Mean fibre length in cm. claculated for 30 days' growth.

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932................	0·791	0·809	0·820	0·824	0·829	0·820	0·814
1933................	0·909	0·944	0·968	0·981	0·954	0·955	0·950

Analysis of Variance.

$l_2 = 1932$ length, $l_3 = 1933$ length.

Sums of Squares and Products.

	l_2^2	$l_2 l_3$	l_3^2
Between groups................	7,866·3	10,769·1	25,635·0
Within groups..................	122,135·4	118,446·8	192,028·6
TOTAL...............	130,001·6	129,215·9	217,663·6

$b = 0 \cdot 9939$.

Analysis of Adjusted Values.

Variance.	Degrees of freedom.	Sums of squares.	Mean squares.
Between groups................	5	11,998·5	2,399·7
Within groups..................	39	74,307·6	1,905·3
TOTAL...............	44	86,306·1	—

$R = 1 \cdot 259$.
$n_1 = 5, \quad n_2 = 39$.

In the case of length, therefore, this insignificant value shows that the variance between groups does not differ from that within groups.

(e) *Mean fibre thickness in μ (weight-length method).*

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932................	15·65	15·85	15·29	15·82	14·93	15·59	15·52
1933................	17·66	17·86	17·13	17·90	17·22	17·56	17·55

Analysis of Variance.

t_2 = 1932 thickness, t_3 = 1933 thickness.

Sums of Squares and Products.

	t_2^2	t_2t_3	t_3^2
Between groups................	4·5405	3·8621	4·0138
Within groups..................	46·4337	42·2156	53·9736
TOTAL...............	50·9742	46·0777	57·9874

$b = 0 \cdot 9094.$

Analysis of Adjusted Values.

Variance.	Degrees of freedom.	Sums of squares.	Mean squares.
Between groups................	5	0·7443	0·1488
Within groups..................	39	15·5721	0·3993
TOTAL...............	44	16·3164	—

$R^{-1} = 2 \cdot 683.$
$n_1 = 39, \quad n_2 = 5.$

Here the same result is obtained as in the case of all the attributes studied, viz., no significant difference between groups.

(f) Mean fibre weight in 10^{-6} gm., calculated for 30 day's growth.

Group.	I.	II.	III.	IV.	V.	VI.	Mean.
1932................	1·98	2·09	1·96	2·09	1·89	1·98	2·00
1933................	2·91	3·07	2·89	3·17	2·90	2·90	2·97

Analysis of Variance.

w_2 = 1932 weight, w_3 = 1933 weight.

Sums of Squares and Products.

	w_2^2	w_2w_3	w_3^2
Between groups................	22·165	29·595	51·901
Within groups..................	359·795	349·533	630·623
TOTAL...............	381·960	379·128	682·524

$b = 0·9715$.

Analysis of Adjusted Values.

Variance.	Degrees of freedom.	Sums of squares.	Mean squares.
Between groups................	5	15·318	3·0636
Within groups..................	39	291·059	7·4630
TOTAL...............	44	306·377	—

$R^{-1} = 2·436$.
$n_1 = 39$, $n_2 = 5$.

Here again as in all the previous cases there is no suggestion of any response to the sulphur supplements, as regards mean fibre weight of the samples.

It will be noted that in all cases the variance between groups does not differ significantly from that within groups. One is therefore led to the conclusion that under the conditions of the experiment, neither the fleece as a whole nor the fibres of a shoulder sample show any indication of having been affected by the supplements.

SUMMARY AND CONCLUSIONS.

(1) Six groups of young wethers were kept on a production ration for two years. During the second year five of the groups received supplements of sulphur and of compounds containing

sulphur, the sixth group acting as control. The grease weights, clean weights, and yield percentages of the fleeces, and the mean fibre lengths, mean fibre thickness, and mean fibre weights of shoulder samples were determined.

(2) The sheep were fed in individual feeding boxes, except for the hay, which was given *ad lib.*

(3) The following supplements were dosed daily, except Sundays:

Group I: No supplement.
Group II: Cystine.
Group III: Sulphates.
Group IV: KCNS.
Group V: 5 gm. sulphur.
Group VI: 0·12 gm. sulphur.

(4) There was no significant difference between the groups, showing that there was no response to any of the supplements, as regards the above-mentioned wool attributes.

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