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## Factors associated with yellowing within Romney fleeces

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### ABSTRACT

Four two-year-old Romney ewes born in the Waikato and four two-year-old Romney ewes born in Canterbury that had been grazed together in the Waikato for 12 months, were used to investigate aspects of yellowing in wool samples grown on the mid-back, mid-side and mid-belly. Two sheep in each sub-group had a history of having a relatively high and two sheep a relatively low, propensity for their fleece to turn yellow during the previous year. Dimensional fibre characteristics, with the exception of staple length, were not significantly different between body sites, between the propensity groups or between birth farm groups. Wool was shorter on the belly than on the other two sites. Incubating greasy sub-samples of each fleece at 40°C and 100% relative humidity for 6 d induced a curvilinear increase in yellowness with the CIE Y – Z value approaching an asymptote. A significant amount of the variation in yellowing was explained by the concentration of K, Ca and possibly Na, elements secreted in the suint. These data suggest that the propensity of a fleece to turn yellow is not related to fleece architecture or the elemental composition of the fibres, but may have a physiological basis possibly related to sweating behaviour.

Keywords: wool; Romney; yellowing; body site; elemental composition.

### INTRODUCTION

Wool yellowing is a complex biological process associated with warmth and moisture (Simpson, 1999). The discolouration may develop while the wool is growing, during storage, processing or useage of the end-product. Some fleeces turn yellow more quickly than others when exposed to the same conditions while wool grown in the lower regions of the body is more yellow than wool grown on the upper regions of the body (Henderson, 1968).

It has been suggested (Hoare & Stewart, 1971; Winder *et al.*, 1998) that under warm, moist conditions, fibres absorb potentially yellow products from water-soluble components of yolk (suint plus wax). Such products tend to accumulate in the lower regions of the body (Dawes, 1973).

A challenge colour test (Aliaga *et al.*, 1996) has been developed to evaluate the potential of wool to discolour under standard temperature and moisture conditions. While there have been several studies on aspects of fleece discolouration between sheep sampled at the same site (Aitken *et al.*, 1994; Bigham *et al.*, 1983; Bray & Smith, 1999) there have been no objective studies comparing variation in discolouration between sites within the same fleece. The trial reported in this paper used the challenge colour test to estimate the magnitude of interrelationships between fleece yellowing, fibre characteristics and elemental content of wool grown on the back, midside and belly for a small group of sheep grazing at one location.

### MATERIALS AND METHODS

#### Sheep

Four two-year-old Romney ewes born in the Waikato on a private farm situated between Kawhia and Raglan and four two-year-old Romney ewes born in Canterbury on a private farm near Ashburton, were used. The sheep were a sub-set of a group of 100 Romney ewes grazed at the Whatawhata Research Centre for the previous 12

months that had been used for a preliminary study of factors associated with wool yellowing. As part of that trial all the sheep were ranked for the propensity of a midside fleece sample to turn yellow under standardised environmental conditions (challenge yellow test – Aliaga *et al.* 1996). Within each of the birth farm (source) groups, two sheep were selected at random from the five sheep which had the highest propensity for their wool to turn yellow (high propensity group), and two sheep were selected at random from the five sheep which had the lowest propensity for their wool to turn yellow (low propensity group), measured over three samplings during the previous year.

The fleece of each of the eight selected sheep was sampled with a shearing handpiece on the mid-back, mid-side and mid-belly regions before shearing a 10-month fleece in December.

#### Wool measurements

Mean staple length of each of the 24 greasy fleece samples was measured by ruler and the total number of crimps along the staple counted. Mean fibre diameter, fibre diameter variation and fibre curvature, of a minicored sample, were measured simultaneously with an OFDA 100 (Edmunds, 1995). Wool colour was described using the CIE system with the derived tristimulus values measured by a Hunterlab D25M reflectance colourimeter. Pre-incubation colour was measured after washing a greasy sub-sample of each fleece in distilled water, drying the sample at 105°C for 3 h and hand-carding it. Post-incubation colour was measured after a second greasy sub-sample from each fleece had been subjected to a modification of Aliaga *et al.*'s (1996) challenge test. Each greasy sub-sample was placed in a 200 ml jar, moistened with deionised water, held at 40°C and 100% relative humidity for 6 d, rinsed in deionised water, left to dry at room temperature and the CIE tristimulus values measured using the New Zealand standard method (NZS 8707:1984).

### Elemental analysis

A weighed sub-sample (~0.5 g) of each of the washed pre- and post-incubation fleece samples was oven dried at 100°C for 12 h, cooled in a desiccator, reweighed and the regain of the sample calculated. A second weighed sub-sample (~0.2 g), weighed at the same time, was digested in concentrated HNO<sub>3</sub> acid, evaporated to dryness and 25 ml 2N HCl added to the residue. The concentration of Na, K, Ca, Mg, P, S, Cu, Zn and Fe were measured by inductively coupled argon plasma emission spectrometry and the concentration adjusted to an oven-dry basis (Lee, 1983).

### Statistical analysis

Individual measurements were analysed by analyses of variance (GENSTAT) (Lawes Agricultural Trust, 1993) fitting effects of source, propensity to turn yellow and body sampling site.

As the concentration of elements that may be associated with wool yellowing could be interrelated, their relative effect on wool yellowing was assessed by multiple regression (GENSTAT, Lawes Agricultural Trust, 1993). The effect of step-wise adjustment to include each element was evaluated based on the change in residual mean square. The critical F-statistic for variate selection was set at  $F = 2$ .

## RESULTS

As there were no significant interactions between source, propensity to turn yellow and sampling site groups for any of the measured characteristics, only the main effects are presented in the following tables.

There were no significant differences between source, propensity to turn yellow or body site effects for total crimps along the staple ( $10.2 \pm 1.3$ ) (mean  $\pm$  standard deviation (SD)), fibre diameter ( $37.2 \pm 2.2\mu\text{m}$ ), fibre diameter standard deviation ( $8.7 \pm 0.9\mu\text{m}$ ) or fibre curvature ( $39.0 \pm 6.6^\circ/\text{mm}$ ). Staple length was also not significantly different between the two propensity groups ( $117 \pm 21\text{mm}$ ) although staples growing on the belly were shorter than staples growing on the midside and back respectively (95 versus 130 and 127 mm; pooled standard error of the difference (SED) = 7mm).

Least-squares treatment means and SED for the tristimulus values of each wool sample in the washed pre- and post-incubation states are given in Table 1. Source effects were not significant for any of the measured tristimulus values. Post-incubation CIE Y – Z was the only tristimulus value to show a significant propensity to turn yellow effect with wool grown by the high propensity group being more yellow (higher CIE Y – Z value) than the low propensity group. Wool grown on the back and midside was not statistically different for any of the measured CIE values for either of the washed pre- or post-incubation samples. The belly wool was however duller in both the washed pre- and post-incubation states (lower CIE X, CIE Y and CIE Z value) and more yellow than equivalent wool samples from either the back or midside of the fleece.

There was a curvilinear response in yellowing during incubation (Figure 1). The yellower the sample was before incubation the less the increase in yellowness during incubation.

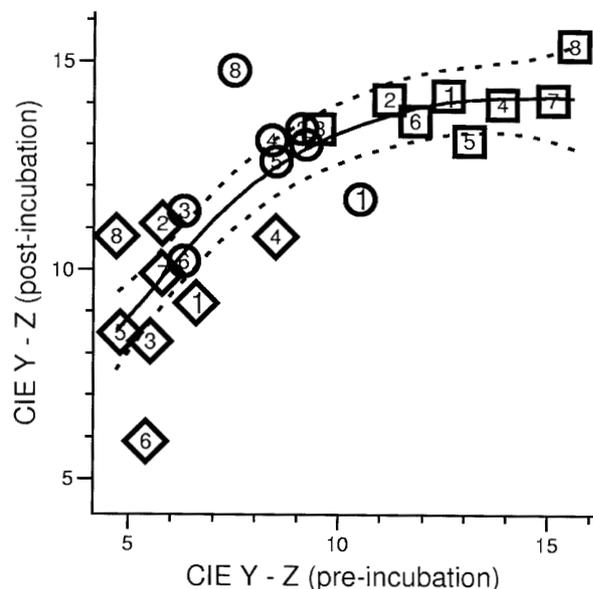
Least-squares treatment means and SED for the elemental content of each pre-incubation wool sample after washing in distilled water are given in Table 2. The only element for which the concentration approached significance between source groups was S, where sheep born in the Waikato had higher values than sheep born in Canterbury. The difference between propensity to turn yellow groups was not significant for any of the measured elements. Site effects varied between elements. Concentrations of Na, Ca, Mg and Fe were higher on the belly than either the back or midside. The concentration of K was higher on the midside than on the back or belly while the concentrations of P, S, Cu and Zn were not significantly different across the three sampling sites.

Least-squares treatment means and SED for the elemental content of each wool sample after incubation in deionised water are given in Table 3. The source effect was not significant for any of the measured elements. Samples from sheep in the low propensity group had higher concentrations of Mg than sheep in the high propensity group. The difference between propensity to turn yellow groups was not significant for any other measured element. Site effects again varied between

TABLE 1: Least-squares means and pooled standard error of the difference (SED) for tristimulus colour measurements of each wool sample in the washed pre-incubation and post-incubation states.

Effect	CIE X		CIE Y		CIE Z		CIE Y – Z	
	Pre-incubation	Post-incubation	Pre-incubation	Post-incubation	Pre-incubation	Post-incubation	Pre-incubation	Post-incubation
Source								
Waikato	48.6	45.0	49.3	43.3	40.3	33.8	9.0	12.0
Canterbury	47.7	46.9	48.2	45.2	39.3	33.4	9.0	11.8
Propensity to turn yellow								
High	47.9	45.9	48.5	44.2	39.0	31.3	9.6	12.9
Low	48.4	46.0	49.0	44.3	40.6	33.3	8.4	11.0
SED	1.3	0.9	1.4	1.0	1.6	1.4	0.8	0.5
Site								
Back	47.8	50.3	48.5	49.0	42.6	39.7	5.9	9.3
Midside	51.8	48.6	52.6	47.0	44.4	34.5	8.2	12.5
Belly	44.9	38.9	45.2	36.8	32.4	22.8	12.9	14.0
SED	1.5	0.3	1.6	0.3	2.0	0.6	0.5	0.3
Source	NS	NS	NS	NS	NS	NS	NS	NS
Propensity	NS	NS	NS	NS	NS	NS	NS	*
Site	**	***	**	***	***	***	***	***

**FIGURE 1:** CIE Y – Z of each sample after being challenged in an incubator, plotted against the CIE Y – Z of the same sample after washing before incubation for samples taken from the back (◇), midside (◻), and belly (□) regions of the same eight sheep. Each sheep is indicated by the numeral 1 to 8. The solid line is the pooled within-sheep relationship across sites and the dotted lines are the 5% confidence limits.



elements. Concentrations of Ca and Mg were higher and the concentration of Zn was lower on the belly than either the back or midside. Conversely, the concentrations of Na, K and Cu were lower and the concentration of S higher on the back than on the midside and belly. The concentration of Fe was higher on the midside than on the back or belly while the concentration of P was not significantly different across the three sampling sites.

The proportion of variation in CIE Y – Z value both pre- and post-incubation, explained by the stepwise inclusion of the concentration of individual elements on the change in residual mean square is given in Table 4. A total of 89% of the variation in CIE Y – Z of the pre-incubation sample was explained by the concentration of Ca, Na, K and Zn whereas 63% of the variation in CIE Y – Z of the incubated sample was explained by K. The inclusion of additional elements had no significant effect in reducing the residual mean square.

### DISCUSSION

Although the relatively small range in fibre dimensions measured in this trial may have restricted our ability to detect statistical significance, the trial results support the work of Bray & Smith (1999) who showed unscourable yellow discolourations within several flocks spread across New Zealand was unrelated to fleece architecture.

**TABLE 2:** Least-squares means and pooled standard error of the difference (SED) for the concentration of Na, K, Ca, Mg, P, S, Cu, Zn and Fe in each wool sample pre-incubation after washing with distilled water.

Effect	Element concentration								
	Na (µg/g)	K (mg/g)	Ca (µg/g)	Mg (µg/g)	P (µg/g)	S (mg/g)	Cu (µg/g)	Zn (µg/g)	Fe (µg/g)
Source									
Waikato	1057	4.03	517	80	138	33.2	5.8	104	80
Canterbury	875	3.76	485	112	129	30.2	5.9	100	41
Propensity to turn yellow									
High	990	4.10	525	87	134	32.1	5.8	106	76
Low	942	3.69	477	104	134	31.2	5.9	98	46
SED	113	0.50	56	18	15	1.2	0.6	11	24
Site									
Back	613	2.29	371	70	135	29.9	6.0	114	51
Midside	955	5.51	337	73	136	32.9	6.0	103	52
Belly	1329	3.89	796	145	131	32.3	5.5	88	80
SED	137	0.82	76	22	2	1.8	0.5	13	4
Source	NS	NS	NS	NS	NS	†	NS	NS	NS
Propensity	NS	NS	NS	NS	NS	NS	NS	NS	NS
Site	**	*	***	*	NS	NS	NS	NS	***

**TABLE 3:** Least-squares means and pooled standard error of the difference (SED) for the concentration of Na, K, Ca, Mg, P, S, Cu, Zn and Fe in each wool sample after incubation with deionised water.

Effect	Element concentration								
	Na (µg/g)	K (mg/g)	Ca (µg/g)	Mg (µg/g)	P (µg/g)	S (mg/g)	Cu (µg/g)	Zn (µg/g)	Fe (µg/g)
Source									
Waikato	370	6.40	617	148	110	28.5	29.3	89	23
Canterbury	428	6.59	531	134	99	28.9	24.2	90	22
Propensity to turn yellow									
High	399	6.79	511	114	104	28.7	25.1	88	22
Low	398	6.20	637	168	105	28.6	28.4	91	23
SED	42	0.85	86	13	8	1.0	3.4	3	3
Site									
Back	233	4.19	397	116	108	30.7	16.2	95	18
Midside	407	7.41	318	65	104	28.2	41.4	93	14
Belly	556	7.88	1006	242	101	27.1	22.6	80	35
SED	52	0.61	52	19	4	0.5	2.7	4	2
Source	NS	NS	NS	NS	NS	NS	NS	NS	NS
Propensity	NS	NS	NS	*	NS	NS	NS	NS	NS
Site	***	***	***	***	NS	*	***	*	***

TABLE 4: Proportion of variance in CIE Y – Z values of washed pre- and post-incubation samples explained by the concentration of individual elements present within the sample.

State	Significant elements	Proportion of variance explained (%)
Pre-incubation	Ca <sup>***</sup> + Na <sup>**</sup> + K <sup>*</sup> + Zn <sup>*</sup>	89
Post-incubation	K <sup>**</sup>	63

The rate of wool yellowing in a growing fleece is known to be dependent on the prevailing environmental conditions. Maintenance of the relative ranking for yellowing over time within this sample of sheep suggests that yellowing is thus influenced by underlying differences in the biology of the fibre.

The positive curvilinear relationship evident in the trial between wool yellowing pre- and post-incubation over the body of the sheep has important implications when ranking sheep for relative propensity of their wool to turn yellow. If wool samples are to be collected for a challenge test they must all be taken from the same body site on each sheep and preferably from a site that has not already developed a pronounced yellow discolouration.

As wool is known to readily absorb a range of dissolved ions from water (R.M.W. Sumner, unpublished data), it was important when measuring the elemental content of wool in this study that only elements bound within the fibre were estimated. Consequently all wool samples were washed or incubated in either distilled or deionised water. Both the use of detergent as a requirement of the colour measurement procedure (NZS 8707: 1984) in the laboratory where the wool was incubated, and the incubation process itself, may each have resulted in the release of more of the weaker bound ions than the washing procedure for pre-incubation colour measurement in another laboratory where detergent was not used. The measured bound ions may thus be considered as either having been incorporated into the fibre matrix during cell proliferation within the wool follicle, or having later diffused into the fibre from the associated wax and suint components in the growing fleece.

The yellow discolouration present in the pre-incubation samples and the yellow discolouration that occurred during incubation did not appear to be visually different. If a particular element were to be directly associated with wool yellowing in the growing fleece it would be expected that the concentration of that element would change between sites that differed in yellowness. It would also be expected that the concentrations of the elements concerned would change differentially during incubation relative to the increase in yellowness. In this trial there was a variable response in the concentration of individual elements that exhibited a significant site effect for wool yellowing pre- and post-incubation. Thus the chemistry of yellowing at different sites in the growing fleece may be different to the chemistry of yellowing that occurs during incubation.

While not evident from analysis of individual elements, regression analyses showed K, Ca and possibly Na concentrations to explain a significant proportion of the variation in yellowing in both the pre- and post-incubation samples. K, Na and Ca are all elements with a high dissociation constant, have few coloured salts and do not readily form complexes. It is therefore more likely, as

postulated by Aitken *et al.* (1994), that their concentration may be indicative of the binding strength of other anions rather than they being a causative factor in their own right.

Suint consists chiefly of K salts of fatty acids (Farnsworth, 1956). Unlike many other animals, the outflow from the sweat glands of sheep and goats is pulsatile with the frequency of the pulses increasing with increasing ambient temperature (Allen & Bligh, 1969). Studies of the physiological factors controlling between-sheep differences in sweating behaviour may provide a possible key to improving our understanding of aspects of the differential rate of wool yellowing between individual sheep in the same flock.

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