THE BILATERAL STRUCTURE OF WOOL CORTEX AND ITS RELATION TO CRIMP

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Summary

New evidence of the nature of the bilateral cortex in crimped wool is presented and discussed in relation to the physical and chemical properties of the wool fibre. The origin of the asymmetry in the cortex was studied by cutting cross sections of sheep skin and swelling plucked wool roots. It is shown that the bilateral structure is present before the cortex is keratinized and does not originate from an asymmetrical keratinization as has been suggested. Coarse wool, in which crimp is much less, are shown to possess a radial asymmetry in which the peripheral cortical cells are less accessible to basic dyes and have a higher sulphur content than the central cells. This is related to the difference between the curved follicles from which fine fibres are produced and the straighter follicles producing coarse fibres.

I. INTRODUCTION

It has been demonstrated that the cortex of a crimped wool fibre comprises two hemi-cylinders differing in both chemical and physical properties, this structural feature bearing a direct relationship to the occurrence of crimp (Horio and Kondo 1953; Mercer 1953b, 1954; Fraser and Rogers 1953, 1954). These differences between the two cortical components can be revealed inter alia by the segmental dyeing produced under controlled conditions, or by numerous keratin swelling or digesting agents applied to intact fibres and fibre cross sections. Horio and Kondo (1953), following the earlier studies of Hirabayashi (1938) and Ohara (1938), confirmed that in wool fibre cross sections one segment was more accessible to basic dyes than the other and this was termed the dye-accessible segment or D.A. This segment was always situated on the outside of the crimp wave and they claimed that the dyeing pattern was reversed with acid dyes.

The present authors prefer the terms H ("hard") and S ("soft"), the S segment being that segment of the cortex which lies on the outside of the crimp wave, and this terminology will be used throughout this paper. H and S refer to properties of the cortex and reflect the differential properties of the segments as related to wool fibre performance, that is, chemical resistance and mechanical strength. It is not intended, a priori, to relate these terms to the established terms of hard and soft keratins used in histological descriptions of the two main keratin types derived from different epidermal tissues. Neverthe-

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less, the differences in properties of the two cortical segments can in part be related to a higher degree of disulphide cross-linking or keratinization.

Experimental results up to the present time suggest that there are two major contributory causes to the appearance of segmentation. One is a fundamental chemical difference in the amino acid composition, particularly in cystine, proline, and dicarboxylic amino acids (Fraser, Lindley, and Rogers 1954) and the other a histological feature, associated with chemically resistant membranes surrounding the cortical cells (Manogue and Moss 1953) and resistant particles in the cells (Mercer 1953a). The resistant particles have been called non-keratinous and have been claimed to exist in greater abundance in the more stable cortical segment (Mercer 1953b). The resistant cortical cell membranes which are of the same order of thickness as the epicuticle have been observed in cross section in the electron microscope (Manogue and Moss 1953) and although there is no evidence for any segmental disposition of these membranes, differences in dye accessibility and resistance to swelling agents may be related to their presence. It is the purpose of the present paper to coordinate and extend published information on this subject and to present a photomicrographic record of some of the most important features of wool-fibre histology interpreted in terms of segmentation and the occurrence of crimp.

II. Experimental

Selected samples of various quality wools and human hair were solvent scoured with 80-100°C b.p. petroleum ether at room temperature, washed several times with distilled water, dehydrated with alcohol, and air dried. Differentiation of the segments was obtained by dyeing with 0·1 per cent. methylene blue in 0·03M phosphate buffer, pH 7·4, at 100°C for 30 min with a wool/liquor ratio of 100 mg/30 ml. A qualitative indication of disulphide bond distribution was obtained by oxidizing with 1·6 per cent. peracetic acid at room temperature for 24 hr with a wool/liquor ratio of 1 g/100 ml and dyeing with 0·0005M methylene blue or toluidine blue at room temperature for 30 min with slight agitation. Cross sections cut in a Hardy microtome were then investigated for dye distribution.

Experiments on cortical segmentation in the follicle were conducted on 8 μ paraffin sections cut from freshly collected sheepskin, showing Merino type crimp, which had been fixed in 5 per cent. formol-saline for 24 hr and treated in the usual way. For staining, the sections were taken down to water and oxidized in 1·6 per cent. peracetic acid for 24 hr. and then thoroughly washed in water. They were then stained in 0·0005M methylene blue (0·1M acetate buffer at pH 2·6) for 2 hr, followed by Mayer’s haemalum and finally eosin as counterstain. Freshly collected wool roots were also treated by several methods before examination.

III. Results and Discussion

(a) Staining Reactions

(i) Basic dyes.—In Merino 64’s quality wool there is a marked difference between the basic dye affinity of the H and S segments as shown in Plate 1,
Figure 1, and under the dyeing conditions specified, uptake of basic dye is practically confined to the S segment and the cuticle. In the S segment there is some differentiation of histological detail as the intercellular cementing material and the contents of the nuclear spaces have dyed more intensely than the cortical cells themselves. In the H segment there is also some dye uptake by the intercellular cementing material and the contents of the nuclear spaces.

The marked difference in basophilia between the segments appears to be localized in the cortical cells, and the boundary between the segments follows the outline of cortical cells. A sharp division of basophilia is apparent and no gradation of properties between the two types of cells has been observed in fibres below about 20-25 μ in diameter. The cross sectional area of the H segment is, in general, somewhat smaller than that of the S segment and the boundary between the segments is parallel to the major axis of the somewhat elliptical cross section. The disposition of the S segment towards the outside of the crimp wave is shown in Plate 1, Figure 2.

(ii) Basic Dyes following Oxidation.—The marked change in the dyeing pattern following oxidation of wool with peracetic acid is of considerable interest. Unlike the staining without oxidation, frequent washing with water did not remove the dye and demonstrated the increased affinity conferred by the oxidation process. Intense staining occurred first in the cuticle and tended to be metachromatic, and maximal staining took place in this structure before cortical staining began. The metachromatic staining was especially obvious with toluidine blue, and was very marked if the oxidized fibres were first swollen with N NH₄OH in order to remove 90 per cent. of the fibre protein and to isolate the cuticular sheaths and cortical cell remnants, and then stained (Plate 1, Fig. 3).

The final staining result with oxidized fibres is shown in Plate 1, Figure 4. It is seen that an intense basophilia has arisen in one cortical segment as well as in the cuticle and that this is more intense than before oxidation. The segment now stained is the H segment (and again is shown to be smaller than the S segment). The identity of the segment has been conclusively demonstrated by carrying out the oxidation with peracetic acid on cross sections of methylene blue dyed fibres whilst observing under the microscope. In this way actual transfer of dye could be observed as it was released from the S segment by the acid reagent and was taken up by the now oxidized H segment.

It is of interest to note that no difference in the segmental staining pattern is observed with the methylene blue solution buffered at pH 7 or at pH 2·6 as employed by Pearse (1951) in his method for staining keratin and adapted by Lillie, Bangie, and Fisher (1954) for the same purpose. These authors did not observe the segmental staining as they used hairs in their investigations.

Mercer (1953b) suggested that segmentation is revealed in cross sections of peracetic acid oxidized fibres if these are swollen in 0·1N NH₄OH, but this is difficult to see in the photomicrographs published by this author and he did not identify the segments. If, however, the peracetic acid treated fibres are first stained as described above, the marked preferential swelling of the unstained S segment produced by 0·1N NH₄OH can be readily followed and
always produces bursting of the cuticle on the S side of the fibre (Plate 3, Fig. 1). This also explains why the action of dilute ammonia on peracetic acid oxidized fibres often leads to longitudinal splitting of the fibre cuticle and liberation of the cuticle as a flat unfolded sheath (Plate 1, Fig. 3).

The reason for the observed increase in basophilia after peracetic acid oxidation is still unknown. Alexander, Fox, and Hudson (1950, 1951) did not find free sulphonic acid groups in completely oxidized wool as evidenced by the inability of this wool to undergo ion-exchange reactions characteristic of a product containing free sulphonic acid groups; however, free cysteic acid was found on acid hydrolysis. Lillie et al. (1954) in their study of this increased basophilia in various keratin structures concluded that sulphonic acid groups were the only ones which could account for the reactions observed, particularly since basic dye staining occurred at low pH values, as observed in the present work.

We found that wool which had been fully reduced with thioglycollic acid and coupled with iodoacetamide did not show any increase in basophilia after treatment with peracetic acid. It seems therefore that at least some free cysteic acid side-chains must be formed in oxidized keratin, and the distribution of the resultant basophilia gives a qualitative indication of the degree of disulphide cross-linking. If this is so, then the results with fine Merino wool indicate that the disulphide cross-linking is greater in the H than the S segment. This observation is further supported by a marked increase in the basophilia of the H segment after the reduction of the disulphide bonds with thioglycollic acid at pH 5.6 followed by coupling with iodoacetic acid (O'Donnell 1954). The increase in the basophilia of the H segment was demonstrated by dyeing with basic dyes at pH 7, but no dyeing occurred at pH 2.6 as with peracetic acid oxidized wool. This is in accord with the difference in the pK values of the sulphonic acid and carboxylic acid groups introduced by these procedures. The distribution of disulphide linkages in the wool fibre cortex revealed by these staining methods has been correlated with sulphur analyses carried out on two fractions obtained from wool by the action of cetyl sulphonic acid in which the segments are separated on the basis of alkali solubility (Fraser, Lindley, and Rogers 1954). The differing physical and chemical properties of the bilaterally constructed cortex are without doubt related in part to this difference in the degree of keratinization.

(iii) Fibre Types and Cortical Segmentation.—Plate 3, Figures 2 and 3, shows the uptake of basic dye in Corriedale 56's quality wool before and after oxidation and Plate 3, Figures 4 and 5, shows that in Lincoln wool. It appears from the study of a large number of cross sections that the disappearance of segmentation, as seen in Merino 64's quality wool, is a function of fibre diameter rather than breed and Plate 3, Figure 3, shows this feature particularly well. In fibres less than about 25 \( \mu \) in diameter the segments are well formed and similar to those of Merino 64's quality.

In the diameter range 25-35 \( \mu \), the sharp demarcation of the segments gradually disappears and the uptake of dye in individual cortical cells is somewhat variable although a classification into H-type cells and S-type cells is
still possible after oxidation. Frequent examples of an H cell surrounded by S cells and an S cell surrounded by H cells are seen in the coarser fibres in Plate 3, Figures 3 and 5. The population of H cells is somewhat less than that of S cells as in the Merino 64’s quality fibres.

Above a diameter of about 35 μ, a division into H and S segments can no longer be justified on the basis of the staining methods described. However, H and S cells may still be differentiated both on the basis of basophilia and disulphide bond content. In the coarser Lincoln fibres of Plate 3, Figures 4 and 5, there is a marked tendency for the H cells to be concentrated around the periphery of the cortex leaving a central core of S cells (cf. Mercer 1952).

The examples of asymmetrical dye uptake discussed above were chosen from a wide range of fibre types as they illustrate the general trends in passing from fine highly crimped wools to coarser wools in which the crimp is much less developed. An additional observation worth recording concerns the “steely” wool produced by sheep maintained on a copper deficient diet, the crimp in this type of wool being very much less than is usual for the particular breed. In the two cases studied ample copper had been restored to the diet of the sheep at a later stage. The later growth was highly crimped, serving as a control. The effect of copper deficiency varied considerably from fibre to fibre; in some cases the segments were ill formed as in coarse wools and in others the difference in basophilia between H cells and S cells was only slight. Similarly anomalous results were obtained after oxidation (Plate 1, Fig. 5).

(b) The Action of Alkalis and Urea

The swelling and breakdown of wool fibres in alkaline media are quite different from that observed in acid media. In general, cortical cells are not released on hydrolysis and the bulk of the protein released appears to be intracellular in origin. A resistant intercellular residue surrounded by a cuticle sheath remnant is commonly observed after alkaline treatments, as for example in Plate 1, Figure 6.

The H and S segments of the cortex differ markedly in their resistance to swelling in dilute alkali. Immersion in 0.1N NaOH for 1-2 hr produces a gross swelling of the S segment with greatly reduced birefringence, whilst the H segment is comparatively unaffected by this treatment. In the presence of thioglycollate the effect is accelerated and is accompanied by the development of blisters on the cuticle around the S segment (Plate 3, Fig. 6).

An electrophoretically pure component extracted from Merino 64’s wool with alkaline thioglycollate solution (Gillespie and Lennox 1953) has been shown to contain more aspartic and glutamic acids and amide N than the intact wool, but less cystine and proline (Simmonds 1955). Since this is believed to be the major protein component of the wool fibre and its removal coincides with a depletion of the S segment, these differences in amino acid composition between component 2 (Gillespie and Lennox 1955) and the whole fibre must reflect similar differences between the H and S segments.

Differentiation similar to that obtained with 0.1N NaOH is obtained by treatment with saturated aqueous urea solutions at 65°C for several days. The addition of a secondary alcohol sulphate-type detergent to the urea solution
accelerates the differentiation and finally the S segment is swollen to about eight times the original volume, whilst the H segment is not visibly affected apart from the development of striations. At this stage the wool fibres may readily be disintegrated by mechanical agitation with glass beads. The disintegrated products include cortical cells derived from the H segment and fragments of cuticle adhering to a jelly like mass which is presumably derived from the swollen S segment.

(c) The Action of Enzymes and Microorganisms

Wool fibres are slowly digested by crude trypsin solutions and certain types of bacteria and fungi with the release of scales and cortical cells. By means of the various methods of differentiating the H and S segments already described it is possible to show that in partly digested wool fibres the S segment is more susceptible to attack and that the first release of cortical cells is from this segment. This is found to be true in crude trypsin digests obtained under sterile conditions, as well as in the case of bacterial and fungal (McQuade, unpublished data) attack on the fibre.

(d) Mechanical Properties

No detailed investigations of the mechanical properties of the H and S segments have so far been attempted, but a number of preliminary observations suggest that they differ to some extent, which is not surprising in view of the gross chemical and histological differences that have been observed. Dr. M. E. Hargreaves, Division of Tribophysics, C.S.I.R.O., attempted the difficult task of performing microhardness tests on thin sections of fine Merino wool. Satisfactory indentations were obtained on a small number of fibres and two depths of indentation were in general obtained on any one section. The ratio of hardness was of the order of 2 : 1. This is attributed to the greater hardness of the H segment. It is hoped to continue this investigation when a more suitable embedding medium has been developed.

A considerable difference in mechanical properties is observed when Merino fibres are extended 50 per cent. in neutral 0·1M thioglycollate solutions. The H segment is highly birefringent whilst the birefringence of the S segment is reduced and accompanied by the development of striations (Plate 4, Fig. 1).

(e) The Action of Acids

When wool fibres are exposed to solutions of low pH, swelling and progressive degradation of the keratin occurs. Under these conditions the H and S segments can be differentiated by differential swelling, an unsymmetrical development of striations, and differential loss of birefringence, indicating a differing susceptibility to acid hydrolysis. Prolonged treatment with dilute mineral acids, or shorter treatment with hydrolytic agents having a catalytic action, result in the release of cortical cells and scales. Again a difference in resistance is observed, indicating that the material of the segments differs considerably.

The differing swelling properties of the bilateral cortex in acid solutions are readily demonstrated by immersing fibres in glacial acetic acid, when a rapid swelling of the S segment is observed, accompanied by a loss of birefringence.
Prolonged treatment with 0·1N HCl or 0·1N oxalic acid at 65°C results in the release of cortical cells. With cetyl sulphonic acid a rapid action is obtained and the two segments are revealed (Plate 2, Fig. 1). Two fractions have been isolated from the wool after treatment with 0·05M cetyl sulphonic acid for 6 days at 65°C (Lindley 1947). One fraction is soluble in dilute alkali and has been shown by microscopical examination after basic dye staining to be derived mainly from the S segment, whilst the insoluble fraction is derived mainly from the H segment and cuticle (Fraser, Lindley, and Rogers 1954).

Amino acid analyses of these fractions have revealed gross difference in the composition of the segments. Thus the resistant fraction is rich in cystine and proline, whereas the alkali-soluble material is lower in cystine but has a higher content of glutamic and aspartic acids. The analytical results are in accord with the segmental staining results discussed above (Section III, (a)), which indicate a high sulphur content in the H segment but a greater basic dye affinity in the S segment. The results can also be correlated with the generally accepted view that the stability of wool keratin towards chemical reagents is largely due to the numerous disulphide linkages which exist both inter- and intramolecularly.

(f) Further Experiments to Reveal Segmentation in Wool Cortex

Four further methods aimed at detecting chemical differences in the two segments as well as differences in accessibility to chemical reagents depending on histological features were unsuccessful. Wool stained with (i) ninhydrin by the method of Cockburn, Jagger, and Speakman (1953), (ii) iodine by the method of Richards and Speakman (1953), and (iii) 2:4 dinitrofluorobenzene by the method of Middlebrook (1951), and studied in cross sections failed to show segmentation. In the last case, reduction of the DNP-wool with stannous chloride followed by coupling with H-acid produced no change in the staining result and so no localized distribution of side-chain -NH₂ groups could be established. Wool stained by immersion for 7 days in 1 per cent. osmic acid buffered in 0·1N veronal at pH 7·4 again showed only a generalized staining of the cortex in cross section.

The action of saturated chlorine or bromine water on wool fibres readily produces segmentation by preferential attack on one of the segments, the action of bromine being the most rapid and destructive to the keratin. It is of interest to note that initially the blister formation (Allwörden reaction) induced by this halogen occurs only on the S side and later spreads all around the fibre surface (Fraser and Rogers 1955). Although this was suggested to indicate differences in the cuticle around the H and S segments (Leveau, Cebe, and Parisot 1953), the most likely explanation is a difference in the molecular size and concentration of the osmotically active substances liberated in the H and the S segments (Fraser and Rogers 1955). This is probably due to differences in the amino acid composition of the two segments.

(g) The Origin of Segmentation

Wool follicles producing crimped wool fibres have a characteristic histological structure which must play a role in the mechanism responsible for crimp
formation. This includes the bulb deflection and bending of the follicle, and the process of asymmetric keratinization of the fibre cortex (Auber 1951). This invariably begins on the thin side of the asymmetric inner root sheath and progresses to the thick side. The cross-sectional dimensions of the outer root sheath on the other hand are variable and take no part in the mechanism. Now that it is known that the wool fibre is bilaterally constructed, it is certain that these factors are responsible for the “setting” of the two segments in a fixed relationship to one another, once they have been formed. Experiments on the swelling and birefringence changes of plucked wool roots induced by various agencies (Plate 2, Fig. 2; Plate 4, Fig. 2) have led to the belief (Fraser and Rogers 1954) that the chemical differences in the proteins of the two segments originate at very early stages of the biosynthesis of the cortex where fibrillation begins and the sulphhydryl reaction can be first detected (Plate 2, Fig. 3). They do not arise as a result of the final hardening process of keratinization which occurs in the lower third of the follicle (Mercer 1954). Furthermore, it seems that keratinization of the S segment may not follow immediately upon its completion in the H segment, these processes occurring in distinct steps rather than continuously across the developing fibre (Plate 2, Fig. 2).

Naturally, it can be suggested that if the bulb deflection is important in the crimping mechanism, then, within one region of the skin where stable crimp exists and fibre crimps are in phase, the follicle bulbs should all be deflected in the same direction. However, this is not found and is not necessarily a prerequisite. Not only are the follicle bulbs at different levels enabling “out of phase” growth, but fibre growth rates are not the same due to follicle-follicle competition, and in any case the staple crimp results from fibre interaction and is not a time indication of “phasing” of the crimp waves from adjacent follicles.

It is now generally accepted that the most chemically resistant segment of the wool cortex (H) is formed on the concave side of the bent follicle, adjacent to the thin side of the inner root sheath. Until recently the evidence for this has been only circumstantial and it has been assumed that the first segment to keratinize should be the H segment as a consequence of its greater degree of disulphide cross-linking. Conclusive evidence that this is the true situation has been presented by Fraser and Rogers (1954) who revealed segmentation in fine Merino wool fibres in cross sections of the wool follicle by the peracetic acid oxidation-basic dye method. It was demonstrated that, almost without exception, the increased basophilia of the H segment was on the thin side of the inner root sheath, and the boundary between the two segments was again situated along the major axis of the elliptical fibre cross section. The presence of the H segment and its disposition in relation to the follicle layers was observed at all levels of the follicle except at the very early stages of fibre formation.

If it is accepted that the action of oxidizing per-acids on wool keratin is to produce side-chain sulphonic groups or at least that the increase in basophilia detected by the basic dye staining at pH 2.6 is specific for acidic groups derived from sulphur-containing side-chains, then such a method should be capable of detecting the level in the wool root at which incipient keratinization takes place. It has been found that when this technique is applied to
sheepskin sections cut at the root level, regions of basophilia, indicating a disulphide content, arise in the presumptive cortex before the morphological changes of keratinization have advanced very far. (Plate 2, Figs. 4 and 5.) Moreover, these regions again appear always on the thin side of the inner root sheath (presumptive H segment) finally spreading until the basophilia completely fills the whole H segment when the fibre has finally keratinized (Plate 2, Fig. 6).

(h) The Origin of Asymmetry in the Cortex of Coarse Wools

With sections of sheepskin from a strong-wool Merino it was found that fibres of large diameter (primary follicles) were often stained in a similar way to that already described for coarse wools. However, the more intense staining tended always to be on the thin side of the inner root sheath as is always the case with the secondary fibres of smaller diameter. In straight-haired animals such as the guinea pig no segmental dyeing was observed, the fibre cortex and cuticle being completely stained and centrally placed in the symmetrical inner root sheath. This is similar to the results obtained by Pearse (1951) in his study of the technique as a “keratin” stain and the later investigations of Lillie et al. (1954).

The differentiation of the H and S segments in Merino fibres is mainly due to the differing dye affinities of the two types of cortical cells rather than areas of the cortex. It is reasonable therefore to discuss the asymmetry of the cortex in coarse fibres in terms of the two types of cortical cell, bearing in mind, however, that histological entities such as cortical cell membranes and intercellular cementing material may also contribute to the dye accessibility of the cortical cells.

H and S cells may still be differentiated in coarser wools but there is a transition from a bilateral structure to one of radial asymmetry as the diameter increases from about 25-40 μ. This transition may be correlated with the change in follicle structure in passing from those producing highly crimped fibres to those producing straighter fibres in which curvature, bulb deflection, asymmetric keratinization, and eccentric disposition of the developing fibre in the inner root sheath are not so marked (Auber 1951).

As mentioned earlier, we have shown that segmental properties exist at the earliest stages of the differentiation of the cortex (Fraser and Rogers 1954). This, together with the clearly defined boundary between the segments suggests that the cell type is predetermined in the germinal layer and does not result from any intra- or extrafollicular influence during development. The course of the metabolism and differentiation of each type of cell, however, is almost certainly related to the asymmetry in the follicle.

In coarser wools the division into two cell types is not so well marked in cross section and the formation of a core of S cells is not readily explained. The cell type may be predetermined in the germinal layer of the follicle, as we have supposed in the case of fine wools, or alternatively the central core of S cells could result from a lesser degree of keratinization towards the centre of the fibre during development. The fact that the cells may still be divided into two types, and the occurrence of isolated H and S cells, however, lends
considerable support to the view that the cell type is predetermined in the germinal layer, and that the amount of crimp is a function of the pattern of their distribution in the developing fibre.

(i) The Isolation of the H Segment

Mercer (1953b) has described a method whereby the S segment of crimped wool, supercontracted in superheated water, can be digested with trypsin, leaving the H segment and cuticle comparatively intact. Analysis of the residue showed that the H segment was considerably richer in cystine than the S segment (Mercer, Golden, and Jeffries 1954), thus confirming the results obtained by Fraser, Lindley, and Rogers (1954). An alternative method (Fraser, Rogers, and Thompson, unpublished data) is to digest the unsymmetrically supercontracted wool with crystalline papain in the presence of urea and bisulphite. The digestion is followed microscopically and when the S segment shows some degradation, the H segment can be isolated by shaking with glass beads and passing the product through a 200 mesh sieve. The isolated H segment, in this case, is not contaminated with cuticle as with Mercer's preparation.

The cystine analyses given by Mercer et al. (1954) are open to the criticism that considerable lanthionine formation occurs during the unsymmetrical supercontraction as evidenced by low solubility in alkaline thioglycollate (Lennox, unpublished data) and paper chromatograms (Thompson, unpublished data). The method described by Fraser, Lindley, and Rogers (1954) is also open to criticism and whilst there is abundant supporting evidence for a preponderance of cystine in the H segment, quantitative analysis of the cystine contents of the H and S segments must await better methods of isolation.

IV. Acknowledgment

The authors are indebted to Dr. M. E. Hargreaves, Division of Tribophysics, C.S.I.R.O., for attempting the difficult task of performing microhardness tests on thin sections of fine Merino wool.

V. References

EXPLANATION OF PLATES 1-4

PLATE 1

Fig. 1.—Cross section of Merino wool fibres stained with methylene blue, showing segmentation and some delineation of cortical cells and nuclear remnants. × 240.

Fig. 2.—Merino wool fibre stained with methylene blue showing S segment on outside of crimp wave. × 240.

Fig. 3.—Cortical cell residues and cuticular sheaths from Merino wool oxidized with peracetic acid, extracted with N NH₄OH and stained with methylene blue. × 240.

Fig. 4.—Merino wool fibres oxidized with peracetic acid and stained with methylene blue. Compared with Plate 1, Figure 1, the H segment is now strongly basophilic. × 240.

Fig. 5.—Irregular staining and segmentation in “steely” Merino wool treated as in Plate 1, Figure 4. × 240.

Fig. 6.—Wool fibre residue following extraction with potassium thioglycollate, pH 12.2, as described by Gillespie and Lennox (1953). Stained with methylene blue. Compare with Plate 1, Figure 3. × 240.

PLATE 2

Fig. 1.—Cross section of cetyl sulphonic acid treated Merino wool fibres stained with toluidine blue. × 240.

Fig. 2.—Freshly plucked wool root swollen in detergent, and photographed in polarized light with gypsum-red plate. The two prekeratinized segments can be seen as distinct entities with birefringence beginning at different levels. × 50.

Fig. 3.—Bulb of plucked wool root stained for sulphydryl groups by the tetrazolium method (Rogers 1953), showing increase in sulphydryl reactivity with development of fibrillation in the cortex. × 240.

Fig. 4.—Section of Merino skin in the follicle bulb region stained by the peracetic acid oxidation-basic dye method. This is at a stage where fibrillation begins but before incipient keratinization and any appearance of asymmetry in the inner root sheath. × 240.

Fig. 5.—Section of Merino skin in the region of incipient keratinization stained as in Plate 2, Figure 4, but showing greatly increased basophilia in one region of the cortex (presumptive H segment) adjacent to the thin side of the inner root sheath. The inner root sheath is not completely keratinized. Unstained regions delineate the cortical cells and stained nuclei are visible. × 540.

Fig. 6.—Section of Merino skin stained as in Plate 2, Figures 4 and 5, and showing two fibres in a late stage of keratinization. Delineation of the two segments is evident, the H segment being the most basophilic and situated on the thin side of the asymmetric inner root sheaths which are almost completely keratinized. × 240.
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PLATE 3

Fig. 1.—Cross section of wool fibres oxidized with peracetic acid, stained with methylene blue, and swollen in 0·1N NH₄OH. The unstained S segment has swollen and the cuticle has burst open. The resistant H segment remains intact and cortical cell remnants are seen in the S segment. × 1175.

Fig. 2.—Cross section of Corriedale 56's quality wool fibres dyed with methylene blue. × 600.

Fig. 3.—Cross section of Corriedale 56's quality wool fibres oxidized with peracetic acid and dyed with methylene blue. × 600.

Fig. 4.—Cross section of Lincoln wool fibres dyed with methylene blue. × 600.

Fig. 5.—Cross section of Lincoln wool fibres oxidized with peracetic acid and dyed with methylene blue. × 600.

Fig. 6.—Appearance of Merino wool fibre during extraction with 0·05M potassium thioglycollate at pH 12·2. The birefringent H segment is on the inside of the curve and the S segment is highly swollen to give an Allwörden reaction-like appearance. × 450.

PLATE 4

Fig. 1.—Merino wool fibre stretched to 50 per cent. extension in 0·1M potassium thioglycollate at pH 7. The two segments can be seen with striations in the S segment. × 3000.

Fig. 2.—Freshly plucked wool root swollen in detergent. The presumptive S segment has dispersed leaving the more stable presumptive H segment. × 250.
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