

STUDIES IN DEPILATION

VII.* STRUCTURAL CHANGES IN THE WOOL FOLLICLE DURING DEPILATION WITH ACID AND ENZYME SYSTEMS

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Summary

Structural changes in the wool follicle during depilation with acetic acid, crystalline trypsin, and a commercial enzyme preparation, Pancrozyme C₁A, are described. A number of the morphological changes produced by acetic acid and Pancrozyme C₁A are similar. Both cause early separation of the epidermal and outer root sheath (ORS) structures from the underlying tissues, and the resistance to fibre withdrawal at this stage is due largely to the physical obstruction of the ORS material. In both cases the separated ORS assumes a tightly packed cylindrical form around the fibre and is gradually digested away until there is no longer any resistance to fibre removal. In both cases separation of the fibre ultimately occurs in the lower part of the prekeratinous zone just above the bulb.

Histological evidence is provided which accounts for the unsatisfactory nature of crystalline trypsin as a depilating agent.

A comparative analysis of the main structural changes in the follicle with the depilating systems at present available is given, and the requirements for an ideal depilating system are stated.

I. INTRODUCTION

It is well established that proteolytic enzymes will cause dermal-epidermal separation, and these agents have been used by many workers for studying various aspects of the epidermis (see Ogura, Knox, and Griffin 1960). For many years enzymes have been suggested for use as depilatory agents and there is an extensive literature on this field (see Green 1952), although their commercial use is not very widespread.

It has also been known for a long time that treatment of skin with acetic acid will cause separation of the epidermis from the dermis (see Baumberger, Suntzeff, and Cowdrey 1942; Ogura, Knox, and Griffin 1960). Lennox (1945) investigated the possibility of using acetic acid to depilate sheepskins, and this was further investigated by Carrie, Moore, and Chisholm (1956). A patent was subsequently taken out (Graham 1957) covering the use of acetic acid as a depilatory for sheepskins.

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In neither of the above cases is the actual mechanism of dermal-epidermal separation or wool loosening understood and it was felt that an histological examination of the structural changes occurring in the follicle during treatment with the above agents would shed some light on the mechanism of the process and ultimately help in the design of a more efficient depilatory system.

The present paper describes the structural changes which take place in the wool follicle during depilation with acetic acid, a commercially available depilatory enzyme (Panerozyme C₁A), and crystalline trypsin.

II. MATERIALS AND METHODS

The sheepskins were selected to be as free from fat as possible. Panerozyme C₁A was obtained from Pancreol Ltd., Halifax, England, and twice-crystallized salt-free trypsin was obtained from Industrial Biochemical Corporation, Ohio, U.S.A.

Samples of skin 3 by 3 in. were taken from a position centrally situated on the backline (position P2, Yates 1965) on the "green" skins. Preliminary work had shown that there were no positional variations in the structural changes taking place. In the case of the acetic acid, a 1M solution of sodium acetate-acetic acid buffer (pH 4.5) was applied to the flesh side of the skin with an inert thickener. The samples were incubated in sealed jars at 28°C. The enzymes were applied to the flesh side of the skin in the form of a slurry at a concentration which contained 4 g of protein nitrogen/litre. In all cases a piece of cheese cloth saturated with the depilatory agent was placed on the flesh side to prevent the agent from flowing off the skin pieces. The samples were incubated in sealed jars at 28°C, and depilation loads* were taken at appropriate intervals as described previously (Yates 1964).

As each depilation load reading was taken, $\frac{1}{2}$ -in. squares were removed from the centre of the sample, processed, and prepared for histological examination following wax embedding as described previously (Yates 1968a).

The histological techniques used for demonstrating the structural features of the follicle were the modified SACPIC method for general structural staining, and the periodic acid-Schiff (PAS) technique for polysaccharides. The details of these techniques have been described previously (Yates 1968a).

III. RESULTS AND DISCUSSION

(a) *Structural Changes in the Wool Follicle Following Treatment with Acetic Acid*

(i) *Depilation Load 17.7 (5 hr)*

There is virtually no breakdown of any of the cells in any component parts of the follicles, and most cells have clearly defined, well-stained nuclei. In spite of the intact cell structure, there are very obvious signs of the epidermis, the outer root sheath (ORS), the bulb, and the glands (Figs. 1 and 2) all separating away from the connective tissue, but there are no vacuolated cells at the points where separation is taking place as is the case with bacterial wool loosening (Yates 1968a). After removal of the fibre the epidermis is unchanged, but the follicle bulb has in many cases been pulled over the papilla and has split (Fig. 3). In a number of cases the bulb has not

* The depilation load is a measure of the force required to remove the fibre from the follicle, and is defined as the force in grams weight required to detach a staple of which 1.5 cm weighs 1 mg. Fresh green skins have a depilation load of c. 80, and a depilation load of 2.0 is regarded as indicating completion of the wool loosening process.

become detached from the papilla but has split about halfway up, the top half being dragged up the follicle.

The inner root sheath (IRS) has undergone some chemical changes and no longer picks up the red stain, but its outline can still be seen.

It is surprising that the depilation load at this stage should be as low as 17·7, as there does not appear to be sufficient separation of the epidermal and ORS structures from the connective tissue sheath (CTS), or general structural breakdown to account for the large drop in depilation load.

(ii) *Depilation Load 14·5 (10 hr)*

The separation of the epidermal, ORS, gland, and bulb structures from the underlying CTS is complete and there is considerable nuclear breakdown. The ORS has contracted towards the fibre (Fig. 4) and its cells are in various stages of breakdown with ill-defined nuclei. As in the case of bacterial depilation, the lower part of the ORS is more degraded than the upper part.

Fibre withdrawal removes the epidermis and in most follicles the bulb becomes detached from the papilla and is pulled up the follicle with the fibre. The remains of the ORS are also drawn up the follicle and the fibre usually breaks off just at the top of the bulb when the physical resistance of the follicle debris becomes too great. Figure 5 shows the point of weakening at the base of the prekeratinous zone of the fibre and the individual cortical cells can be seen. The broken-off bulb and the agglomeration of ORS debris at the top of the follicle are seen in Figure 6. This picture is confirmed by the appearance of the lower half of the follicles which are quite empty, and the appearance of the root ends which all show traces of bulb material.

(iii) *Depilation Load 7·6 (21 hr)*

The nuclear staining has completely disappeared, except for a very faint positive response in the cells of the glands. It is of interest that the outlines of the gland cells can also still be seen and it is apparent that these cells are more resistant to destruction and degeneration than the other cells of the follicle. The ORS is completely separated at all points from the CTS and has the appearance of a thin cylinder around the fibre (Fig. 7). The separation of the bulb material from the papilla, which has been observed to a small degree in earlier sections, is complete by this stage. The follicle bulb, in spite of being completely denucleated, has maintained its overall integrity and when the fibre is removed the bulb is dragged up the follicle and becomes detached from the fibre in the upper half of the follicle, forming some of the follicle debris. The remains of the ORS and the fibre bulb can be seen puckered up in the top part of the follicle and forming a physical barrier to easy removal of the fibre, and transverse sections show this mass to be completely amorphous and non-nucleated (Fig. 8).

A few of the root ends at this depilation level are tapered but there is still a majority with varying degrees of follicle material attached, including some with the entire bulb.

Red-staining IRS material is completely absent, but in a number of the transverse sections before fibre removal there is a glass-like ring of dark green-staining material surrounding the fibre in the position where the IRS should be.

(iv) *Depilation Load 4.9 (28 hr)*

The cells of the glands have become disorganized and have lost their outline, and their nuclei no longer stain. The ORS is further constricted around the fibre, and when the fibre is removed the remains are again seen puckered up at the top of the follicle. More of the root ends of the fibres are becoming tapered but there are still considerable numbers that are removed with follicle material attached.

(v) *Depilation Load 1.2 (38 hr)*

All the degenerative processes discussed previously are complete. After removal of the fibre the follicles still contain some amorphous debris, but degeneration has proceeded to the extent that there is much less puckering of ORS and bulb material at the top of the follicle when the fibre is withdrawn. Even at this stage quite a number of fibres are withdrawn with some bulb material attached.

The basement membrane is still quite intact and undamaged at the end of the process (Fig. 9).

(b) *Structural Changes in the Follicle following Application of the Depilatory Enzyme, Pancrozyme C_{1A}*

(i) *Depilation Load 21.9 (5 hr)*

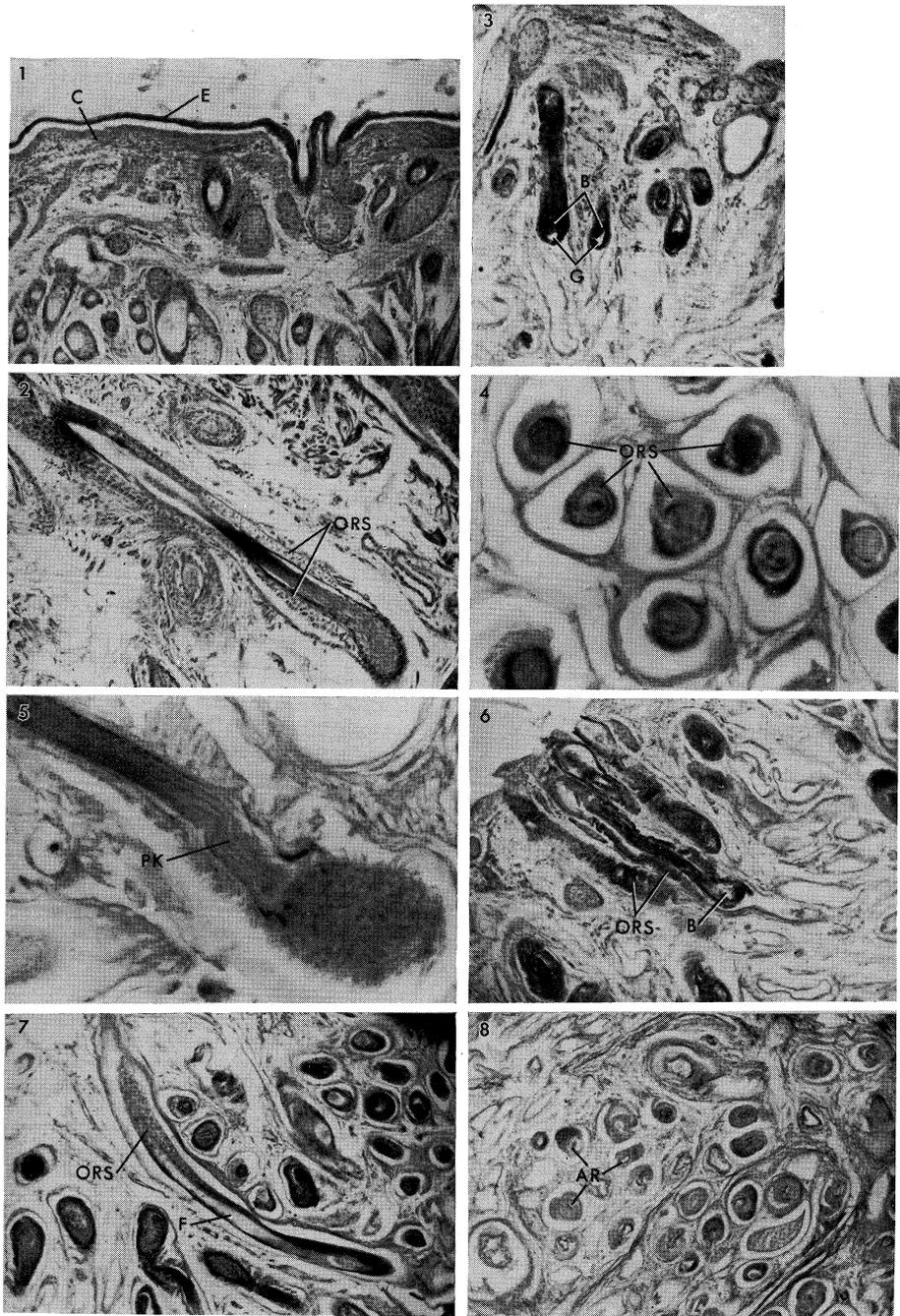
The most outstanding feature at this early stage is the complete separation of the epidermis, the ORS, the follicle bulb, and the sebaceous glands from the underlying connective tissue (Fig. 10) without, however, any cell breakdown or diminution in the intensity of nuclear staining. The ORS, forming a cylinder around the fibre (Fig. 11), is very similar in appearance to that observed after the acetic acid treatment. It is difficult to tell whether the ORS has become slightly compacted around the fibre or if the outer layers of cells have dissolved. Transverse sections show that the IRS is still intact.

Other workers (Einbinder, Walzer, and Mandl 1966) have observed that the first effect of treatment of skin with proteolytic enzymes is vacuolation and perinuclear halo formation in the cells of the basal layers, followed by dermal-epidermal separation. With Pancrozyme C_{1A} no changes were observed at this stage in the

Figs. 4-6.—Acetic acid treatment, depilation load 14.5. **4**, High power transverse section showing contraction of ORS around the fibre, and the denucleated state of the ORS cells. **5**, High power section of the prekeratinous zone (*PK*) showing point of fibre weakening and the individual cortical cells. **6**, Separated bulb (*B*) and the puckered remains of the ORS material in the upper half of the follicle after fibre removal.

Fig. 7.—Acetic acid treatment, depilation load 10.1. Shows the ORS in the form of a tightly compacted tube around the fibres (*F*).

Fig. 8.—Acetic acid treatment, depilation load 7.6. Amorphous, non-nucleated remains (*AR*) of the ORS after fibre removal.



Figs. 1-3.—Acetic acid treatment, depilation load 17·7. **1**, Separation of the epidermis (*E*) from the underlying collagenous structures (corium, *C*). **2**, Separation of the outer root sheath (*ORS*) from the corium. **3**, Shows follicle bulb (*B*) after detachment from the papilla and displacement up the follicle. *G*, gap where the papilla used to be.

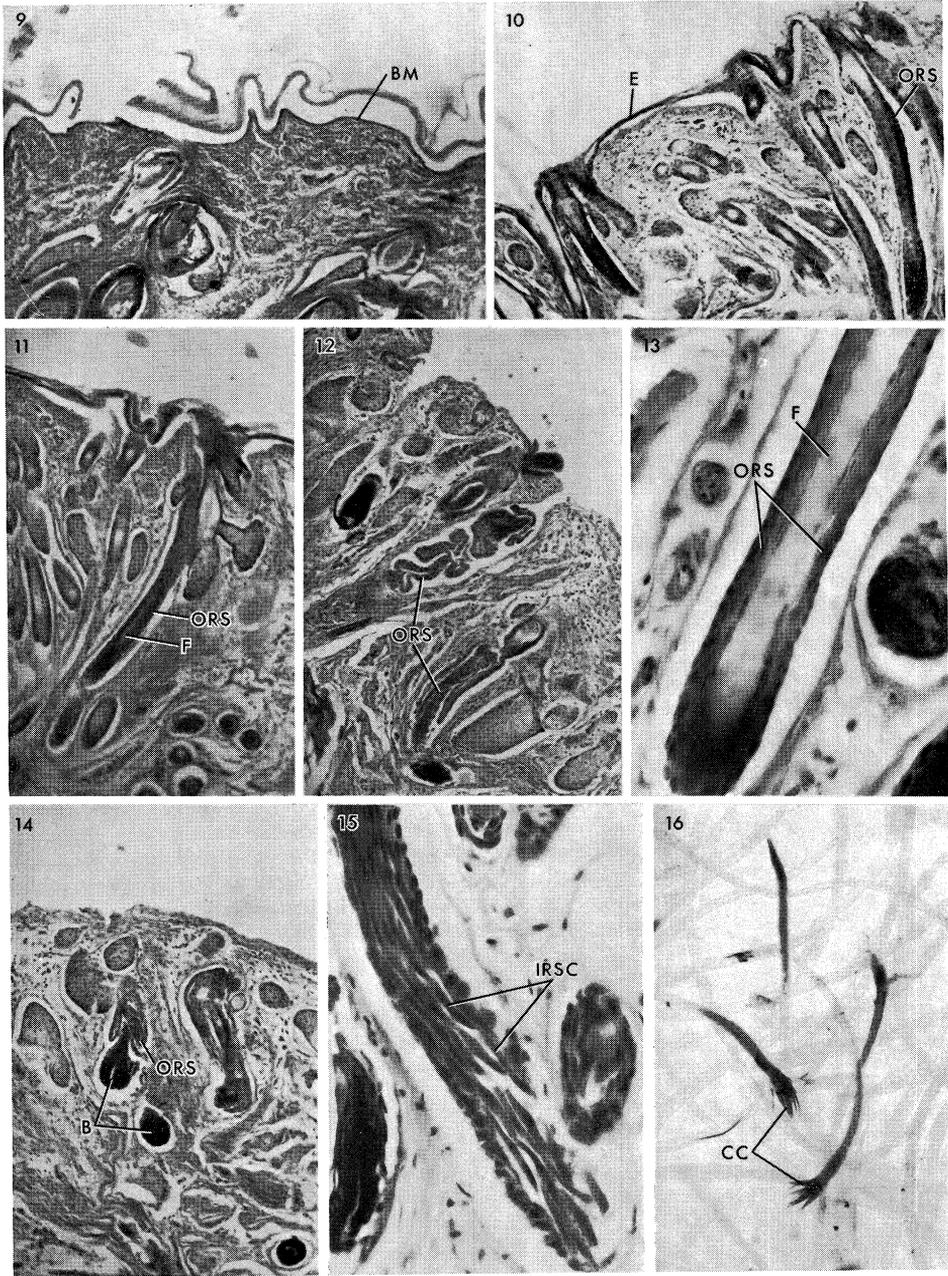


Fig. 9.—Acetic acid treatment, depilation load 1·2. Shows PAS-positive basement membrane (*BM*) still intact underneath the epidermis at the end of the depilation process.

Figs. 10–12.—Pancrozyme C₁A treatment, depilation load 21·9. **10**, Separation of the epidermis (*E*) and the ORS showing the absence of any cell or nuclear breakdown. **11**, ORS forming a compact cylinder around the fibre (*F*). **12**, Puckered remains of the ORS (fully nucleated) at the top of the follicle after fibre removal.

cells of the basal layer, but with the bacterial method of wool loosening ("sweating") (Yates 1968a) dermal-epidermal separation was preceded by vacuolation of the cells as described by Einbinder, Walzer, and Mandl (1966).

If the fibre is removed at this stage the entire epidermis is removed, the intact IRS is withdrawn with the fibre, and many of the fibres have filamentous ends. Much of the ORS debris remains in the follicle and in many cases the puckered remains of the ORS are seen at the top of the follicle (Fig. 12). The bulb remains in position when the fibre is removed and the fibre breaks off at the base of the prekeratinous zone. It appears that insufficient cell destruction has taken place to permit the depilation load to drop to an acceptable level, in spite of the complete separation of the epidermal structure.

(ii) *Depilation Load 9.9 (7 hr)*

The ORS can be seen as a more compacted cylinder around the fibre, and the nuclei have a flattened appearance as though the cells are under a compressive force (Fig. 13). The drop in depilation load between this and the last stage can probably be accounted for by disappearance, or merely a softening, of some of the ORS material, permitting easier withdrawal. When the fibre is withdrawn the follicle bulb is pulled over the papilla and is usually pulled up the follicle until the resistance of the remaining ORS material causes the fibre to break. The puckered remains of the ORS and the bulb can be seen in Figure 14, and the similarity of the changes to those produced by treatment with acetic acid (Fig. 6) is marked, the main difference being that in this case the nuclei still stain intensely. Most of the fibres have filamentous ends when removed and have the IRS still attached.

(iii) *Depilation Load 4.9 (10 hr)*

The ORS appears to be more compacted around the fibre, but there is very little actual cell disintegration in the lower part, as was the case with the acetic acid-treated sample. The changes on fibre removal are the same as those described in the above section with the exception that the degree of puckering of the ORS material at the top of the follicle is less. The IRS is showing signs of disintegration into the characteristic spindle-shaped cells (Fig. 15) when the fibre is removed. The cell structure of the sebaceous glands is quite intact at this stage, again indicating that it is more resistant to the damaging effect of enzymes.

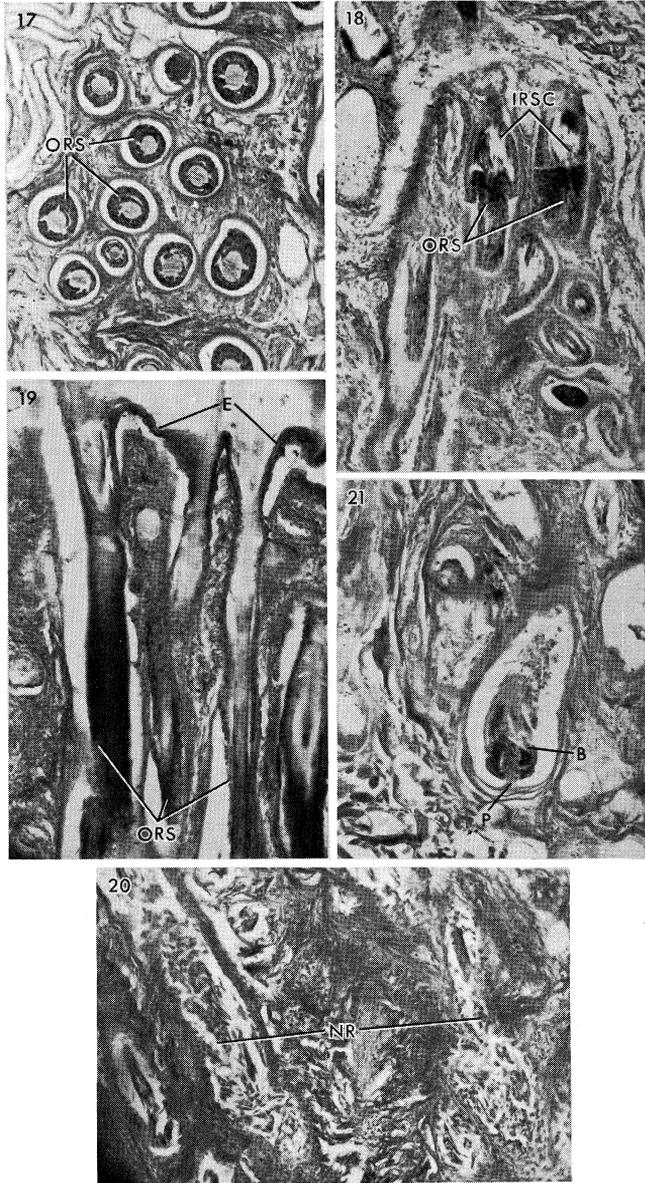
(iv) *Depilation Load 1.7 (14 hr)*

Even at this stage the nuclei are quite intact and have not lost any intensity of staining. The IRS is starting to fragment even with the fibre in position and is

Figs. 13 and 14.—Pancrozyme C₁A treatment, depilation load 9.9. **13**, High power view of compacted ORS around the fibre (*F*), showing the flattened cell nuclei. **14**, Follicle bulb (*B*) after separation from the papilla and detachment from the fibre. Note the puckered ORS remains, and the intact state of the cell nuclei compared to the acetic acid treatment (Fig. 6).

Fig. 15.—Pancrozyme C₁A treatment, depilation load 4.9. Disintegration of the IRS into characteristic spindle-shaped cells. *IRSC*, inner root sheath cells.

Fig. 16.—Pancrozyme C₁A treatment, depilation load 1.7. Root ends of fibres withdrawn at the completion of depilation, showing the filamentous cortical cells (*CC*).



Figs. 17 and 18.—Trypsin treatment, depilation load 61·2. **17**, Separation of fully nucleated ORS from the underlying connective tissue, and apparent contraction around the fibre. **18**, Removal of fibre showing puckering of the ORS remains and early fragmentation of the IRS into spindle-shaped cells (*IRSC*).

Fig. 19.—Trypsin treatment, depilation load 30·5. Complete separation of the epidermis (*E*) and the ORS from underlying connective tissue.

Fig. 20.—Trypsin treatment, depilation load 6·3. Nucleated, fragmental remains (*NR*) of the ORS after fibre removal.

Fig. 21.—Trypsin treatment, depilation load 1·3. Follicle bulb (*B*) still firmly attached to the papilla (*P*) at the conclusion of the depilation process.

completely fragmented when the fibre is withdrawn. Fibres withdrawn at this stage show no adhering IRS material and all have filamentous ends (Fig. 16).

The ORS is still present in a tight cylinder around the fibre and the cells are very much flattened and have elongated nuclei. Only the lower end of the ORS and the prekeratinous zone are showing signs of disintegration, but it is a very different type of disintegration from that seen in bacterial wool loosening (sweating) where there are no cellular remains or nuclei — here there are both. Another point of difference to the sweating process is that with the Pancrozyme C₁A the ORS is removed uniformly all along the fibre, whereas in the sweating method cell disruption commences at the bottom of the ORS and slowly moves up towards the epidermis. When the fibre is removed some follicles still show puckering of the ORS in the top of the follicle, but this is not as marked as at higher depilation loads, presumably because there is less of the ORS material left. In many follicles the ORS material is not disrupted by fibre removal, perhaps because the IRS has largely disintegrated, leaving a gap between the fibre and the ORS. In all cases the fully nucleated bulb remains behind in the follicle.

The basement membrane is completely intact at the end of the enzyme process.

(c) Structural Changes in the Wool Follicle following Application of Crystalline Trypsin

Trypsin has been shown (Yates 1968c) to be a poor depilatory enzyme, and an investigation of the histological changes during trypsin loosening may throw some light on the reasons for this.

(i) Depilation Load 61.2 (10 hr)

Many of the changes seen in sections after this time are those associated with a much greater drop in depilation load in other processes, e.g. the bacterial method, and the use of Pancrozyme C₁A. The epidermis has begun to separate and in many parts is quite free of the corium (cf. Pancrozyme C₁A). In the areas where separation has occurred, the basal cells are in many cases vacuolated and have pycnotic nuclei (cf. sweating). This latter observation agrees with that of Einbinder, Walzer, and Mandl (1966) although no acantholysis was observed in these experiments as was reported in Einbinder's work. The lower part of the ORS has separated from the CTS (Fig. 17) and there has been considerable disintegration of the ORS cells at this level (cf. sweating). In many cases the upper part of the ORS also has separated, but not to the same extent as the Pancrozyme sample. The IRS is in many cases fragmenting into the characteristically shaped cells. This may be expected as it is known that trypsin readily dissolves IRS proteins (Rogers 1962). Removal of the fibre completely removes the epidermis (cf. Pancrozyme), and produces a certain amount of puckering of the undissolved ORS material and fragmentation of the IRS into cells (Fig. 18); both of these phenomena are associated with a much lower depilation load in the sweating and Pancrozyme procedures. The fibre breaks off just above the bulb and on removal leaves most of the bulb material, often in a disrupted condition, *in situ*. On removal fibres show no attached IRS material and have filamentous or, in a few cases, tapered ends.

(ii) *Depilation Load 30.5 (21 hr)*

The main change is in the separation of the epidermis and the ORS from the underlying tissues, which is almost complete (Fig. 19), showing a remarkable resemblance to the Pancrozyme samples at a much lower depilation load. There has been no diminution in the intensity of nuclear staining. Fibre removal produces puckering of the ORS, and in some cases inversion, especially where separation of the ORS is not complete at the top of the follicle.

(iii) *Depilation Load 6.3 (43 hr)*

Disintegration of the epidermis, the ORS, and the gland cells is advanced, and there is complete fragmentation of the IRS into the spindle-shaped cells. The remains of the ORS in the follicle (Fig. 20) are unlike those seen in the sweating and the Pancrozyme treatments. In this case, the ORS seems to break down into nucleated fragments, a stage not seen in the other two methods. The puckering of the ORS on fibre removal is not seen due to this fragmentation. The general appearance of the sections resemble those treated with Pancrozyme, but it is interesting that in no case has the bulb become detached from the papilla, and fibre removal frequently disrupts the bulb *in situ*.

(iv) *Depilation Load 1.3 (59 hr)*

Even after this time, all the nuclei are still intensely staining, although a number are pycnotic. The ORS, IRS, and epidermis are all completely broken down into fully nucleated cells. Prolonged exposure to the trypsin has still failed to detach the bulb from the papilla, which is left in position when the fibre is removed (Fig. 21). This is in direct contrast to the changes in the sweating and Pancrozyme techniques where the bulb is always separated from the papilla at some stage. The basement membrane is quite intact and undamaged at the end of the process.

It is apparent that much more actual cellular disintegration of the follicle components occurs before the fibre is completely loose in this case than with either the sweating or Pancrozyme treatments. It appears that at all stages of depilation with trypsin the fibre breaks off just above the bulb at the base of the prekeratinous zone, but histological evidence suggests that the prekeratinous zone is not weakened as rapidly with trypsin as, for example, with Pancrozyme. Combination of this with the fact that at all stages the follicle bulb remains tightly bound to the papilla means that the depilation process becomes protracted.

It is a feature of the trypsin-treated skins that there is a much greater variation in the state of the follicles at a particular stage than is seen with any other depilatory treatment. The changes described above are those for the most advanced state of degradation at each depilatory level, but especially in the early stages many follicles had not reached the stage of degradation described. It is of interest in this context that Einbinder, Walzer, and Mandl (1966) find that skins must be incubated in enzyme solution for consistent dermal and epidermal changes to occur, and that topical application of enzymes gives inconsistent results. This variability in the trypsin system is undoubtedly a contributing factor to the unsuitability of trypsin as a depilatory enzyme.

There have been conflicting reports on the ability of crystalline trypsin to effect separation of the epidermis from the dermis. Medawar (1953) and Cooper (1958) found that purified trypsin did not bring about dermal-epidermal separation, while a commercial trypsin did. Medawar attributed this to the presence of elastase in the commercial sample, which was regarded as essential for dermal-epidermal separation. However, Fan (1958) states that crystalline trypsin was active in this respect, and indeed dermal-epidermal separation was observed in the present work, although a longer time was required than with another proteolytic enzyme system, Pancrozyme C₁A.

IV. GENERAL DISCUSSION AND CONCLUSIONS

One of the most significant observations in all the methods of depilation described in this and previous papers (Yates 1968*a*, 1968*b*) is that the PAS-positive-staining basement membrane remains structurally and chemically intact throughout the process as judged by its staining characteristics. The entire depilation process takes place outside this layer and the chemical reagents responsible for depilation have to move through it to the site of action. The ability of the basement membrane to retain its integrity under various forms of stress has been commented on by previous workers (Burbach 1959; Pearson and Spargo 1961), and the present work provides additional evidence for the structural and chemical toughness of this membrane.

It is clear that the dermal-epidermal junction is one of the key sites of depilatory action, and one of the main objectives of depilation (other than with methods using reducing agents which have a direct effect on the keratin) is to destroy the adhesion of the outer root sheath cells, and incidentally the epidermal cells, to the basement membrane.

The wool loosening process involves two stages, which may occur concurrently. There is first of all the separation of the outer root sheath and usually the epidermal structures from the basement membrane, and secondly a certain amount of actual cell destruction is required. It may be possible to get some idea of the relative parts played by these two stages in the overall process by considering depilation with acetic acid, where the depilation load drops from a level of *c.* 80 to 17 in 5 hr, and depilation with Pancrozyme C₁A, where the depilation load falls to 21.9 in 5 hr, in both cases as a result of separation and without any actual cell destruction. Completion of the loosening process takes place by cell destruction. Both these methods of depilation are quite rapid and it is significant that in the bacterial sweating process, where appreciable separation does not take place until substantial cell destruction has occurred, depilation is a much slower process. It is also significant that in the case of depilation with ammonia where both stages occur simultaneously, although not to a satisfactory level in either case, there is a very rapid large drop in the depilation load (Yates 1968*b*).

Sections of skin depilated with acetic acid, ammonia, and Pancrozyme C₁A all show an agglomeration of outer root sheath material in the top part of the follicle when the fibre is removed in the early stages of depilation. This material causes a physical obstruction to the removal of the fibre and keeps the depilation load at a high level. In the case of acetic acid and Pancrozyme C₁A, destruction of the outer

root sheath cells proceeds to the extent that the material remaining does not provide any resistance to fibre removal and the depilation load drops to an acceptable level. The gradual diminution of the amount of outer root sheath material left to obstruct fibre removal as depilation proceeds can quite clearly be seen from the sections. In the case of the ammonia depilation, however, the process of cell destruction reaches a certain level and then stops, which partially accounts for the failure of the depilation load to drop to an acceptable level.

Although separation and partial destruction of the epidermis occur in depilation, it is unlikely that this is an integral part of the depilation process and it is likely that its separation occurs simultaneously or, in some cases, later than that of the outer root sheath simply because its basal cells are attached to the underlying structures in the same manner as those of the outer root sheath.

The overall evidence suggests that destruction of the IRS is not essential for satisfactory wool loosening to occur and is only an incidental part of the depilation process. In many cases the IRS appears intact and can be removed in its intact state surrounding the fibre at the end of depilation. In other cases the IRS fragments into spindle-shaped cells only at the end of the depilation process. A strong piece of evidence which supports the lack of involvement of the IRS is that in the case of trypsin, which is a poor depilatory enzyme, the IRS is destroyed at a very early stage while the depilation load is still virtually unchanged.

The changing pattern of the root ends as depilation proceeds sheds some light on the mechanism of the process, although the actual pattern varies considerably from method to method. With the methods using enzymes (including bacterial sweating), where the fibres are withdrawn at some stage prior to the completion of depilation, most of the fibres show quite clearly separated cortical cells at the end of the fibres. This indicates that the intercellular material in the prekeratinous zone is being dissolved as a consequence of which the fibre is weakened at this point. In nearly all cases fibre breakage in the early stages of depilation occurs at the base of the prekeratinous zone and the fibre is removed without any bulb material. Obviously at this stage the attachment of the bulb to the papilla is offering more resistance to removal than is the breaking load of the prekeratinous zone. Towards the end of the depilation process the fibre ends become more tapered as the enzyme or the chemical action completely destroys the integrity of the prekeratinous zone almost up to the level of keratinization — this is seen quite clearly in the staining characteristics of the fibre. In the case of depilation with acetic acid, however, the fibre bulb is rapidly loosened to the extent that the fibre is mostly withdrawn with some bulb material attached, and only towards the end of the process are the filamentous cortical cells seen. In the case of depilation with ammonia, softening of the prekeratinous zone does not occur to the extent where individual cortical cells can be seen, and the fibre breaks off at the junction of the lower end of the prekeratinous zone and the top of the bulb, where considerable cell destruction of the bulb cells can be seen.

It is of interest that in the cases where depilation does not proceed to completion as in the case of ammonia treatment, or where it is very protracted as in the case of trypsin, the follicle bulb has not become detached from the papilla. Even in the sweating method, where depilation is a relatively slow process, the follicle bulb is seldom detached from the papilla, but rather degrades *in situ*.

In conclusion, it may be said that an ideal rapidly acting depilatory would have the following properties in probable order of importance. It would be able to quickly loosen the attachment of the bulb to the papilla (cf. acetic acid), it would be able to quickly effect the separation of the outer root sheath from the underlying structure (cf. Pancrozyme C₁A), and it would be able to cause sufficient cell destruction of the ORS to permit easy withdrawal of the fibre.

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