

STUDIES ON THE DEPILATORY ACTIVITY OF SODIUM SULPHIDE AND SOME RELATED COMPOUNDS

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Summary

The depilatory action of sodium sulphide on sheepskin has been investigated by measuring the pull required to remove wool from skin, the swelling of wool root shafts, and the mechanical strength of wool. The rate of action is augmented by increase in temperature, pH value, and concentration of sulphide in the solution employed up to 1.0M, but is reduced by increase to higher concentrations. The reciprocal action of the hydroxyl and hydrosulphide ions in digesting keratin is explained in Appendix I in terms of the ionization of sulphydryl groups.

Solutions of 1.0M sodium sulphide, partially oxidized with peroxide and adjusted to pH 12.8, completely loosen wool in 18 hours at 20°C. only if the oxidation-reduction potential is lower than -400 millivolts. Salts of various oxyacids of sulphur are less effective depilatories than sodium sulphide, the activity diminishing with increase in the number of oxygen atoms in the anion and approximately with the heat of formation.

Sodium selenide and sodium telluride possess depilatory activity equal to that of sodium sulphide.

I. INTRODUCTION

For hundreds of years, chemicals have been used in the leather industry to remove hair, wool, and epidermis from skins and hides prior to tanning. Lime, alone or "sharpened" with arsenic sulphide, was used originally, but recently sodium sulphide was found to have pronounced depilatory powers and it is today the most widely used adjunct to lime. Merrill (1925), Marriott (1928), and others have determined the optimal conditions for the un-hairing of hides. The findings of these workers are not directly applicable to the problem of de-wooling sheepskins for, whereas hair is of so little value that in the tannery the usual procedure is to immerse the hides completely in the un-hairing solution, wool is such a valuable product that precautions must be taken to prevent damage by the alkaline solutions used to loosen it from the skin. Consequently, the technique of applying the depilatory as a paste to the flesh side of the skin has been developed to minimize contamination of the wool.

This chemical de-wooling process has received practically no scientific investigation in the past and it is still largely empirical. The viscosity of the paste, the concentration of sodium sulphide, and the time of application vary from place to place and, although the concentration is sometimes varied according to the thickness of the skins, strict control is seldom exercised. Usually,

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too, the wool is removed approximately 24 hours after application of the depilatory to the skins, regardless of the hastening or retarding influences of such factors as temperature and thickness of skin.

If the depilatory has low viscosity, and is therefore easily applied, excess may contaminate the wool at the edge of the skin and weaken or even dissolve it. Contamination also occurs when the wool is pushed off the skin onto the depilatory-contaminated pulling beam. In addition, the calcium compounds in this contaminated wool and on the root ends of the detached wool precipitate calcium soaps during scouring and contribute further to deterioration in the quality of the wool. Consequently, wool obtained by chemical loosening may be characterized by a harsh handle, lack of lustre, and the presence of particles of calcium salts, disintegrated wool, and weakened fibres, which break up and are lost during combing.

It was with the object of defining more accurately the minimum requirements for depilatory activity, with special reference to the influence of physical and chemical factors, that the present investigation was undertaken.

II. METHODS OF INVESTIGATION

(a) Preparation of Reagents

Except when otherwise stated, the reagents used were of analytical reagent quality or better. The sodium salts of hydrogen sulphide, selenide, and telluride were specially prepared.

Sodium sulphide was prepared from the chemically pure salt, hydrogen sulphide being liberated by mixing it with hot hydrochloric acid, washed by bubbling through water, and absorbed in pure sodium hydroxide. This solution of sodium hydrosulphide was stored in sealed paraffin wax-lined bottles and diluted with water and adjusted to the required pH with sodium hydroxide immediately before use. Although, at the pH values used in these investigations, the material was actually a mixture of sodium sulphide and hydrosulphide, for convenience it is referred to as sodium sulphide and its properties are accurately defined by the pH and the molarity in terms of the hydrogen sulphide content.

Sodium selenide and sodium telluride were prepared immediately before use by direct combination of the elements at an elevated temperature in the absence of oxygen (Vallance, Twiss, and Russell 1931). The exact stoichiometric weights of the elements were used to avoid the formation of polycompounds. Sodium selenide was also prepared by reduction of sodium selenate with carbon, and by combination of the elements in the vapour phase at low pressure.

The kieselguhr and kaolin employed were finely ground commercial mineral specimens.

(b) Preparation of Depilatories

The chemical substances used in the depilatories were dissolved in distilled water. The actual content of active reducing material was determined by standard iodometric procedures (Hall 1936). Unless otherwise stated, the

solutions were adjusted to a suitable physical state with kieselguhr. Before use, the pH of the depilatory was adjusted to the required value by the addition of hydrochloric acid or sodium hydroxide. For pH measurements, a Leeds and Northrup valve potentiometer was used in combination with a Cambridge high alkalinity glass electrode.

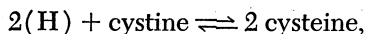
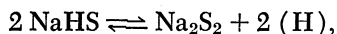
(c) *Measurement of Wool Loosening Activity*

The method of measuring wool loosening was that due to Lennox (1945). From the central area of a fresh sheep skin, pieces 5×5 cm. were cut, and blood and dirt removed by thorough rinsing in running water. In most experiments the skin pieces were then immersed for 30 min. in an alkaline buffer of the same pH as the depilatory to be applied. Sodium carbonate (0.2M) or 0.2M trisodium phosphate was used for this purpose.

A measured amount of the depilatory was applied to the flesh side of the skin, covered with a glass plate to restrict evaporation and oxidation, and the whole then placed in a moist chamber at 20°C. The progress of wool loosening was followed by the determination, at suitable time intervals, of the depilation load. A depilation load of less than 1 was considered to indicate complete loosening of the wool.

(d) *Measurement of Eh*

Any chemical process that depends for its efficiency on reduction must be influenced by the reducing intensity of the system. The normal measure of this, particularly in biological systems, is the oxidation-reduction potential, but this only applies if the system is reversible. In the system now under study there are the reversible reactions:



and the irreversible oxidation of sulphide to sulphite or thiosulphate.

The irreversible nature of the latter reaction and the ease with which the platinum electrodes are poisoned by systems containing sulphydryl compounds render Eh measurements difficult and of doubtful accuracy. However, some measurements were made using a valve potentiometer and platinum electrodes similar to those described by Gillespie (1946). The electrodes were cleaned in boiling concentrated nitric acid after each reading, a mean value from four electrodes being obtained for each Eh. The potentials were converted to the H_2 standard and the results given later are presented in this form.

(e) *Fibre Strength Measurements*

Histological observation has shown that most de-wooling chemicals act by reducing the strength of the keratinous region of the wool root, and consequently the mode of action of depilatories can be investigated by determining their effect on wool fibres.

The single fibre breaking-strength apparatus described by Fookes (1946) was used in these investigations. The technique is open to the criticism that

no provision was made for constant loading rates or for maintaining a humidified atmosphere. These were overcome by care in operation and the inclusion of samples of wool of known strength at frequent intervals during testing. Observations on these revealed any change in testing conditions. With these precautions, satisfactory comparative figures were obtained.

The chemicals under test were dissolved in air-free distilled water, sometimes with the addition of the wetting agent, cetyl trimethylammonium bromide. The wool used was merino 60-64's cleaned by extraction with solvents in the following order: high aromatic light petroleum, acetone, water, acetone. About 30 kg. of wool was thus treated and after careful picking to remove foreign matter, it was thoroughly randomized. It had a normal content of cystine and possessed no detectable sulphydryl groups.

(f) *Wool Root Studies*

The wool fibres with roots attached were pulled from a sheep each day. The roots, consisting of root bulb with about 5 mm. of attached shaft, were placed in the cavity of a well slide, a few drops of the reagent under test was added, and the preparation covered with a slip and placed on the stage of a dissecting microscope. Swelling was detected and semi-quantitatively measured with an eyepiece micrometer. There was usually a lag period before swelling commenced, followed by a period of rapid expansion lasting about 30 sec., and finally a much longer period during which the rate of swelling became progressively slower. The recorded time of swelling was that required for the completion of the lag and rapid phases.

III. EXPERIMENTAL RESULTS

(a) *Influence of Various Conditions on the Depilatory Activity of Sodium Sulphide*

(i) *pH*.—Four procedures were used to determine the pH at which optimum wool loosening occurs. In the first, duplicate skin pieces were soaked in buffers over the range pH 9 to 13 and then were treated with sufficient depilatory, containing 1.0M sodium sulphide at the same pH as the buffer used, to give a standard weight per unit area of skin. Depilation loads were measured at $\frac{1}{2}$, 1, 2, 4, and 18 hours at 20°C. The results at 18 hours are plotted in Figure 1A. Variation in the thickness of skin pieces caused errors in the above determinations. The second method eliminated these errors. A whole sheepskin was treated with a 1.0M sodium sulphide depilatory, and after 24 hours, depilation loads and surface pH values were determined at a number of positions on the skin. The results of this experiment are given in Figure 1B.

The third method demonstrated a relationship between the pH of solutions of sodium sulphide and the rate of swelling of the shafts of wool roots immersed therein. This gave an indication of the conditions required for initial weakening of the wool root shaft in the follicle. The results are given in Figure 2. In the fourth method, the relation between pH of a sodium sulphide solution

and its effect on the breaking strength of wool fibres was determined, the concentration being maintained constant by the use of a very large ratio of solution to wool. These results are shown in Figure 3.

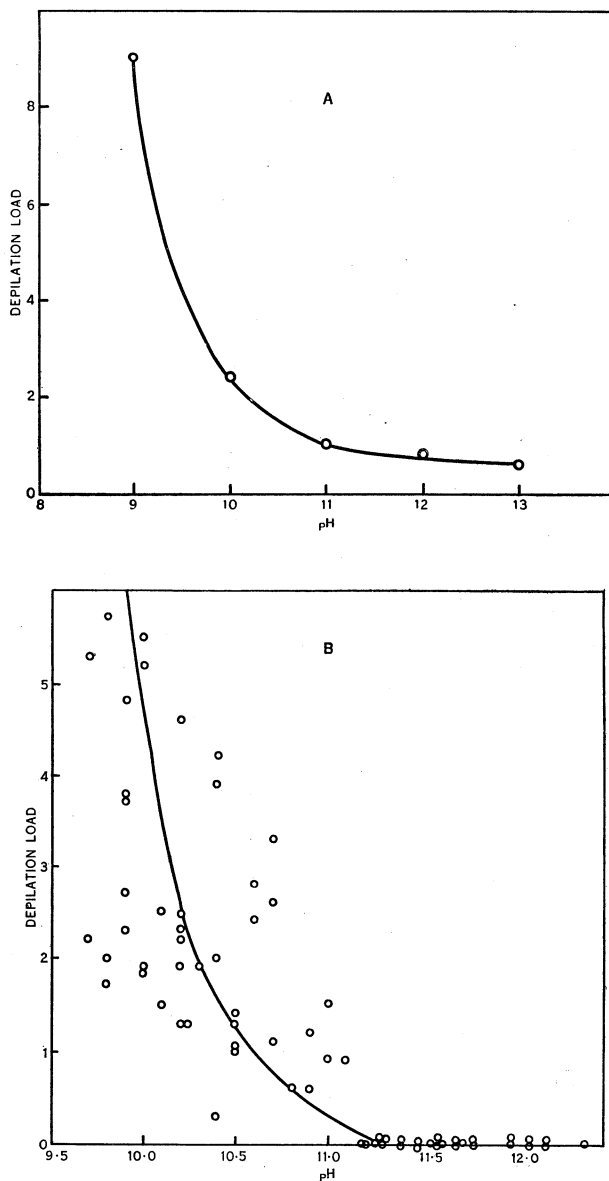


Fig. 1.—A. Relationship between pH of 1.0M sodium sulphide solutions and depilation load.

B. Relationship between pH of 1.0M sodium sulphide solutions and depilation load. (Determined by measuring local surface pH values and corresponding depilation loads.)

It is interesting to observe the profound influence of pH on the extent of wool loosening and wool damage. The approximate effective values of pH are tabulated in Table 1. An explanation of the differences in pH value will be suggested later.

TABLE 1
MINIMUM pH VALUES AT WHICH 1.0M SODIUM SULPHIDE EXERTED A SIGNIFICANT EFFECT ON WOOL ROOTS AND WOOL FIBRES

Reaction	pH at which Reaction is Extensive
Wool root shaft swelling	9.5
Wool loosening	11.0
Reduction in strength of wool fibres	11.5

(ii) *Concentration of Sodium Sulphide*.—Two methods were used to determine the effect of concentration of sodium sulphide on the depilatory action. In the first method, several skin pieces were soaked in buffers at five pH values

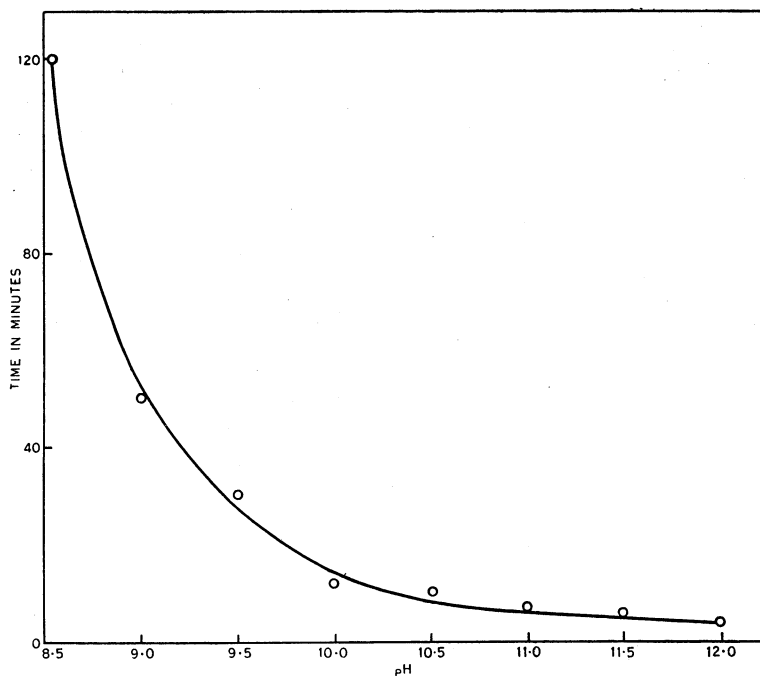


Fig. 2.—Influence of pH on the time taken for 1.0M sodium sulphide to cause visible swelling of the wool root shaft.

and then treated with depilatories at the same pH values but containing concentrations of sodium sulphide covering the range of 0.01 to 1.0M. In Figure 4 the depilation loads for such a series of skin pieces cut from one skin are plotted against the logarithms of the sodium sulphide concentration. In some experiments, the concentration of sodium sulphide was increased beyond the usual

1.0M. It was then observed that instead of increasing further, the activity actually decreased (Fig. 5). These results could be reproduced by adding sodium chloride to a 1.0M solution of sodium sulphide. It therefore seems probable that the osmotic effect of high salt concentrations opposed the depilatory effect of the sulphide. In the second method, reported in Figure 6, the relationship between sodium sulphide concentration and the breaking strength of wool fibres at six different pH values was determined.

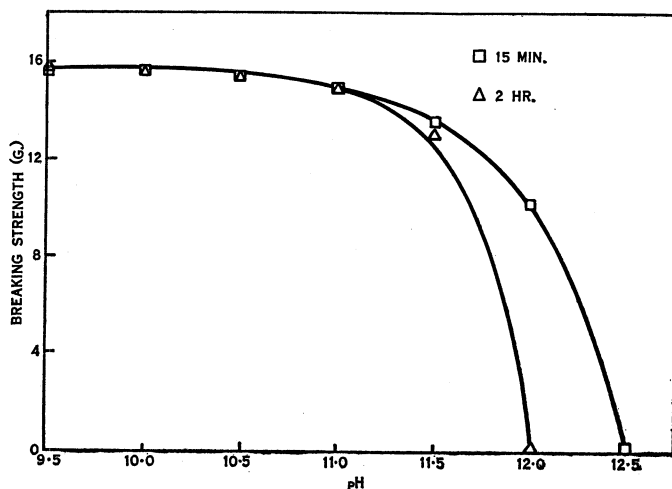


Fig. 3.—Relationship between the pH of 1.0M sodium sulphide solutions and their ability to lower the breaking strength of the wool.

It is apparent that there is an interrelation between pH and the concentration of sodium sulphide solution as measured by either wool loosening activity or the extent of wool damage. The most noteworthy point is the effectiveness of very weak solutions at the higher pH values. Thus 0.025M sodium sulphide at pH 13 is as effective as 0.25M solution at pH 12 and 0.75M solution at pH 11.5, whilst even in 1.0M concentration, solutions at pH 11.0 and 10.5 could not reduce the breaking strength to zero in the time allowed. They did so, however, over longer periods.

(iii) *Oxidation-Reduction Potential*.—A series of nine aliquots of 1.0M sodium sulphide were partially oxidized by nine hydrogen peroxide solutions having graded concentrations such that the most concentrated was just sufficient to completely oxidize the sulphide. A tenth aliquot of sodium sulphide was used in the completely unoxidized state. The pH was maintained at 12.5 during the oxidation by the occasional addition of NaOH.

The ten solutions were thickened with kieselguhr and the pH adjusted to 12.8. Oxidation-reduction potential measurements were made on a portion of each and the remainder was applied in duplicate to skin pieces. The depilation load was measured after 18 hours at 20°C. Figure 7 shows that wool loosening became detectable at potentials more negative than -300 millivolts and

complete loosening was accomplished at potentials more negative than -400 millivolts.

(iv) *Temperature and Time*.—As would be expected, depilation load is also a function of temperature and time of action of the depilatory. The curves in Figure 8 were obtained when skin pieces that had been preheated to the temperature of the experiment were treated with $0.5M$ sodium sulphide depilatory adjusted to pH 12.8 and preheated to the same temperature as the skin to which it was applied. The temperature was maintained constant during the experiment and depilation loads were measured at suitable intervals of time. The higher the temperature, the more rapidly the depilation load diminished. The upper limit was, of course, set by the low shrinkage temperature of the skin in the presence of $1.0M$ sodium sulphide, which is $53^{\circ}C$. (Lennox, unpublished data).

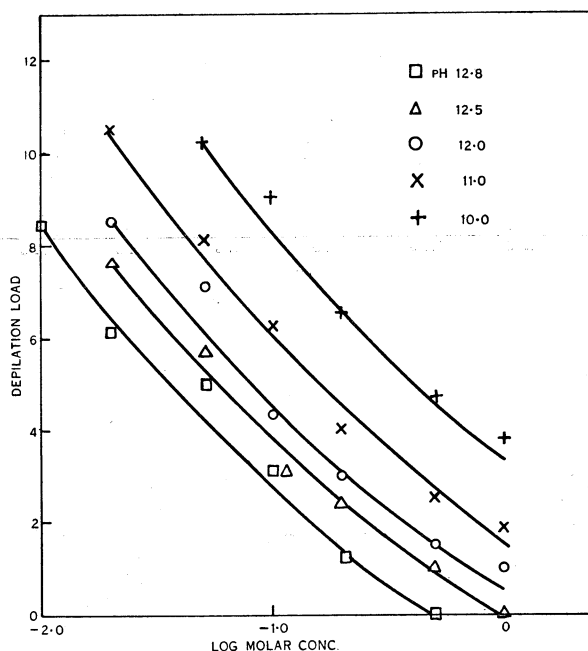


Fig. 4.—Relationship between concentration of sodium sulphide and depilation load after 18 hours at various pH values at $20^{\circ}C$.

(b) *Depilatory Activity of Various Inorganic Compounds Containing Sulphur in the Anion*

Depilatories at pH 12.8 containing the usual amount of kieselguhr but with the sodium sulphide replaced by sodium salts containing sulphite, sulphate, thiosulphate, dithionite, dithionate, or trithionate ions, respectively, were tested in the usual way on buffered skin pieces. The results are given in Table 2, together with the heats of formation of the acids from which the salts were

derived (Hodgman and Holmes 1945). It will be noted that, when arranged in order of increasing heat of formation, which is related to reducing capacity,

TABLE 2
DEPILATORY ACTIVITY OF SOME COMPOUNDS CONTAINING SULPHUR IN THE ANION,
TESTED AT 1.0M CONCENTRATION AND pH 12.8

Compound	Formula	Depilation Load After 18 hr.	Heat of Formation of the Corresponding Acid (kcal.)
Sodium hydroxide control	NaOH	36	—
Sodium dithionate	$\text{Na}_2\text{S}_2\text{O}_6$	33	274.31
Sodium tetrathionate	$\text{Na}_2\text{S}_4\text{O}_6$	33	262.37
Sodium trithionate	$\text{Na}_2\text{S}_3\text{O}_6$	33	261.89
Sodium sulphate	Na_2SO_4	34	210.28 (dil.)
Sodium dithionite	$\text{Na}_2\text{S}_2\text{O}_4$	8	166.1 (dil. sol.)
Sodium sulphite	Na_2SO_3	6	145.09 (300)
Sodium thiosulphate	$\text{Na}_2\text{S}_2\text{O}_3$	6	138.6 (1,500)
Sodium sulphide	Na_2S	1	9.56 (liquid)

a series is obtained within each group of compounds resembling that obtained by arranging them in order of increasing depilation load.

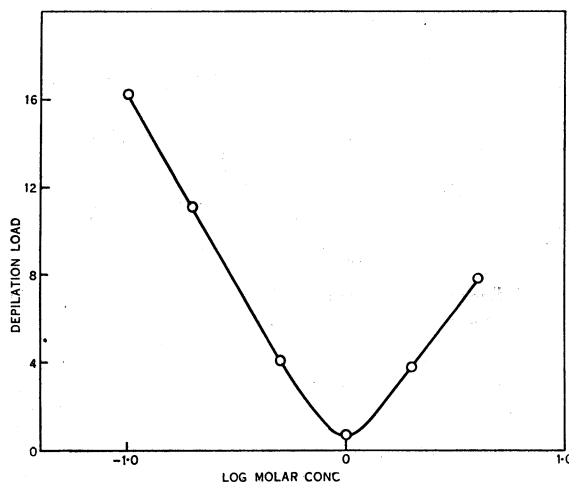


Fig. 5.—Relationship between concentration of sodium sulphide and depilation load showing the inhibitory effect of high concentrations. The samples were incubated for 18 hours at 20°C. before measuring the depilation load; pH was held at 12.5.

Sodium polysulphides up to Na_2S_4 were also prepared and tested and, in general, they showed a decrease in depilatory activity with increase in S content. However, in some experiments it was observed that a mixture of 0.9M sodium sulphide and 0.1M Na_2S_2 showed greater depilatory activity than pure sodium sulphide.

The Australian fellmongering industry relies on an impure commercial grade of sodium sulphide for preparing depilatories. This is usually a fused product containing as impurities polysulphides, carbonates, sulphites, thiosulphates, and sulphates, Al and Si in appreciable amounts, and traces of B, Cu, Ca, and Ga. When depilatories of identical pH, ionic strength, and SH con-

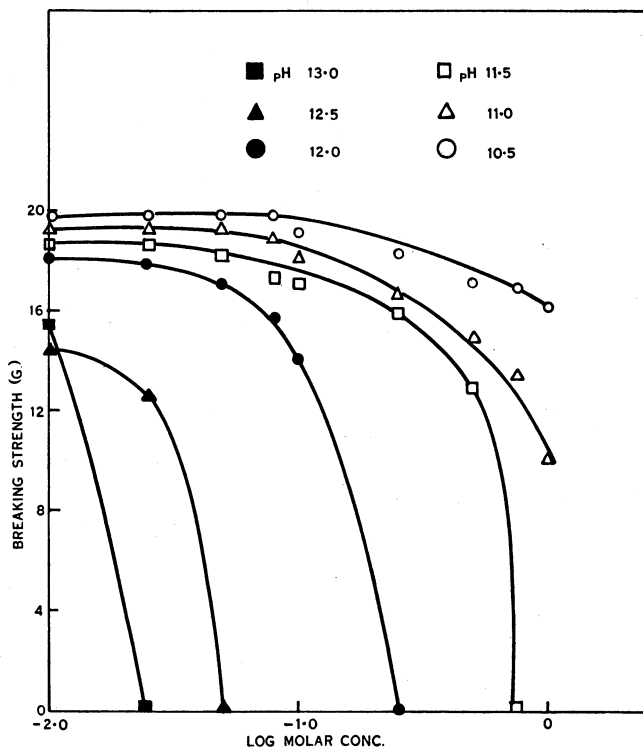


Fig. 6.—Influence of sodium sulphide concentration at various pH values on the tensile strength of wool after 30 min. at 20°C.

tent were tested side by side, one prepared from pure sodium sulphide and the other from crude sodium sulphide, the latter had the greater activity. A typical set of results is shown in Table 3. The effect was most noticeable with

TABLE 3
THE GREATER DEPILATORY ACTIVITY OF IMPURE THAN PURE SODIUM SULPHIDE AT 20°C.

Depilatory	Depilation Load			
	0 hr.	½ hr.	2 hr.	18 hr.
Sodium sulphide 1.0M	58.1	34.3	8.5	0.5
Sodium sulphide 1.0M (crude)	57.5	1.0	0	0

thick skins that were hard to depilate, presumably owing to their high fat content and thick collagen layer.

By fractional crystallization the active fraction was concentrated in the dark brown mother liquor, and the addition of a small quantity of this concentrate to pure sodium sulphide exerted a marked activating effect. The mother liquor was rich in polysulphides but its activating effect could not be reproduced by synthetic polysulphides prepared by dissolving sulphur in sodium sulphide.

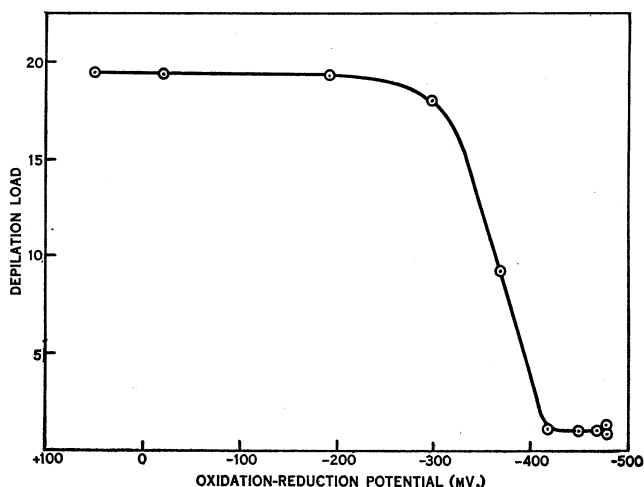


Fig. 7.—Influence of oxidation-reduction potential on the rate of wool loosening by oxidized 1.0M sodium sulphide solutions at pH 12.8 after 18 hours at 20°C.

(c) *Depilatory Activity of some Compounds of Selenium and Tellurium*

In an attempt to determine what impurities in commercial sodium sulphide are responsible for its high depilatory activity (because of their own rapid depilatory action or by catalysis of the action of sodium sulphide) an examination was made of some of the impurities that are most likely to promote activity. These were considered to be other sulphur compounds and substances containing the related elements, selenium and tellurium. When investigating this latter possibility, it was observed that the alkali and alkaline earth metal salts of hydrogen selenide and telluride in suitable concentration and at pH's greater than 11 were very active as depilatories. Some of the results are shown in Table 4.

Under the conditions of the experiment, sodium selenide and telluride appear to have much the same pH threshold as sulphide and possess approximately the same depilatory activity as sulphide when compared on a molar basis.

IV. DISCUSSION

Some divergence can be observed between the minimum pH values for the action of sodium sulphide as determined by various methods. Thus, rapid swelling of wool root shafts occurred at a lower pH than depilation and this, in turn, was lower than the pH associated with measurable reduction in the

strength of wool fibres. A possible reason is that swelling of the wool root is only an early stage in the weakening of the root that is necessary for depilation, whilst the more drastic treatment required for wool fibre damage may be

TABLE 4
DEPILATORY ACTIVITY OF 1.0M SOLUTIONS OF SODIUM SULPHIDE AND SODIUM TELLURIDE AT pH 13, AND OF SODIUM SELENIDE AT VARIOUS pH VALUES, AT 20°C.

Time (hr.)	Depilation Load					
	Na ₂ S at pH 13	Na ₂ Se at the Following pH Values:				Na ₂ Te at pH 13
		13	12.5	12	11.5	
0	32	29	35	35	34	30
½	34	23	39	35	34	33
1	17	16	19	23	24	20
2	9	8	7	9	10	8
18	0	0	0	2	3	0

due to the resistant nature of this material as compared with the newly formed keratin of the wool root shaft. Below pH 10 depilation is difficult to achieve, even with concentrated solutions of reducing agents, and this is probably associated with a corresponding difficulty in completely reducing -S-S- bonds below this pH value (Patterson *et al.* 1941).

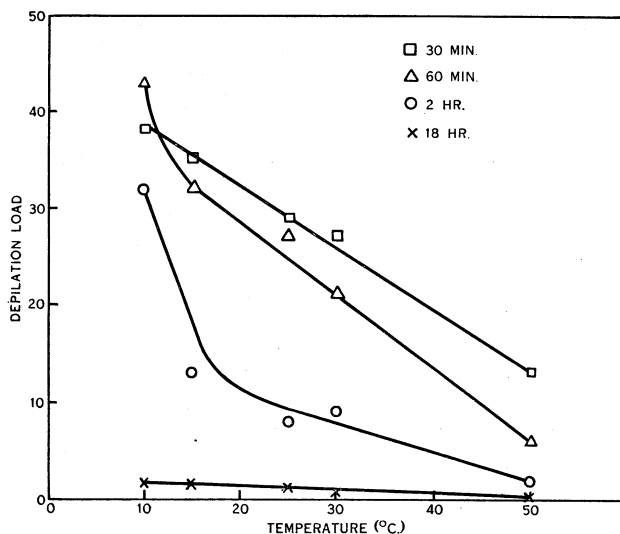


Fig. 8.—Influence of temperature on the rate of wool loosening by 1.0M sodium sulphide at pH 12.8.

Inhibition of depilation by high concentrations of sodium sulphide or by sodium sulphide plus sodium chloride was observed on many occasions. The exact cause is probably not simple, but two possibilities suggest themselves.

The first, which lacks experimental evidence, is that, by withdrawing water from the skin tissues, penetration of the depilatory chemicals is retarded. This is in keeping with the observation that depilation is impossible to achieve with dry, or partly dry, skins. The second suggestion is that sodium salts interfere with the attack on keratin. Vago (1937) has stated that an essential part of keratin lysis is reduction of $-S-S-$ bonds and then splitting of the keratin bundles by absorption of water. It has been observed that high concentrations of salts inhibit the swelling of wool root shafts in dilute sodium sulphide solutions and the second explanation therefore seems to be the more likely.

From the experimental data available it is impossible to state an optimal temperature for depilation of sheepskins, as the rate of depilation increases with rise in temperature until the skin shrinkage temperature is reached. It would be expected that increasing the temperature would accelerate diffusion of chemicals through the skin, partly of its own accord and partly by increasing the rate of hydrolysis of fatty layers that hinder penetration. The reaction of sulphide and hydroxyl ions with the wool root shafts would also be accelerated. There may be a temperature at which there is considerable keratin lysis but little damage to other skin proteins, but this has yet to be revealed. Similarly it is impossible to specify at this stage any particular concentration and pH value of sodium sulphide for effective depilation, for the one is dependent on the other, as shown in Appendix I.

TABLE 5

INFLUENCE OF CONCENTRATION AND TIME ON THE pH REACHED IN THE CENTRE LAYERS OF SKIN PIECES IMMERSSED IN SODIUM CARBONATE SOLUTIONS AT 20°C.

Molar Concentration	pH of Solution	pH in Centre Layer of Skin			
		15 Min.	30 Min.	60 Min.	120 Min.
0.1	11.5	—	9.6	10.0	10.3
0.2	11.8	—	9.9	10.1	10.5
0.5	12.2	—	10.0	10.5	11.3
1.0	12.3	9.8	9.9	10.7	11.7

Since depilatories are not applied directly to the wool root shaft but to the flesh side of the skin, the active components OH^- and SH^- must pass through the skin before reaching their site of action. No appreciable reaction takes place between the SH^- and the skin proteins, and a high level of SH^- is found within a few minutes at the epidermal level. However, the skin proteins react with appreciable quantities of OH^- and there is always a marked pH gradient from the flesh side to the wool side. In Table 5 some figures for the penetration of OH^- are given. The pH values were measured by inserting a high alkalinity glass electrode into a cavity formed by slitting the skin, and were confirmed by using indicators.

It is evident that the depilatory not only has to provide OH^- for the reaction with keratin but also has to bring the pH of the skin proteins up to 10-11. In many experiments it has been found that sodium sulphide solutions alone (1-1.5M) do not provide sufficient OH^- , although there is a great excess

of SH^- . It is now clear why lime has proved such a useful thickener for sodium sulphide depilatories, for it provides an adequate reserve of OH^- which at no time can attain a sufficiently high concentration to damage the pelt.

The use of lime has many disadvantages but alternatives are difficult to suggest. The provision in the skin of an initial reserve of OH^- by immersing it in a solution of Na_2CO_3 provides one successful alternative and a second is to be found in the provision in the depilatory itself of extra OH^- by the controlled addition of NaOH .

On theoretical grounds it was anticipated that Na_2Se and Na_2Te would be active depilatories and that there would be an increasing activity up the series Na_2S , Na_2Se , and Na_2Te as evidenced by the relative ease of oxidation of these compounds. Failure to detect a corresponding increase in wool loosening activity might be due to partial oxidation during preparation and application of the depilatories.

V. ACKNOWLEDGMENTS

Dr. S. J. Leach and Mr. E. F. Woods have provided helpful contributions to the theoretical aspects of Appendix I. The authors are also grateful to Miss C. M. M. Mackinnon and Mr. H. R. Day for their technical assistance.

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APPENDIX I

THE RECIPROCAL ACTION OF HYDROXYL AND SULPHYDRYL IONS IN THE
DIGESTION OF KERATIN

By W. G. CREWTER* and J. M. GILLESPIE*

A physicochemical treatment of the reduction of the dithiol linkages of wool by alkali sulphides is made difficult firstly, by the heterogeneous nature of the system, and secondly, by our lack of exact knowledge of the reaction taking place. However, the experiments described in this appendix suggest that, over a limited range of pH values, a simple relationship exists between the pH and sulphide concentration of solutions bringing about a constant degree of wool degradation. It is considered therefore that the theoretical treatment here presented is of use in suggesting further experiments, though it cannot be considered rigorous.

It has been suggested by Preisler (1930) that reactions between thiol compounds do not approach a true equilibrium. However, the experiments on which this opinion was based were carried out at very low pH values and it has been shown by Ghosh, Raychaudhuri, and Ganguli (1932) that, at pH values exceeding 7.0, thiol-dithiol systems behave as redox systems in the absence of oxygen. As the experiments to be described were conducted at pH values exceeding 9.0, it has been assumed that a true equilibrium is approached and that the usual electrode equations can be used to describe the system at equilibrium. In view of the inherent inaccuracies in the experimental method, activity coefficients have been neglected.

The relationship between the limiting values of pH and sulphide concentration necessary for reduction of the tensile strength of wool to zero was determined as follows:

A solution of potassium sulphide, prepared from A.R. potassium hydroxide and washed hydrogen sulphide gas in air-free solution, was suitably diluted with air-free water and aliquots of each dilution were adjusted to various pH values. These solutions were numbered in a random manner prior to setting up the experiments, and aliquots were taken for sulphide estimations by the usual iodometric technique (Hall 1936), the solutions being acidified after mixing with the iodine solution. A 20 ml. portion of the sulphide solution and 0.1 g. of well-mixed solvent-scoured wool were used for each test, and the digestion was carried out in an atmosphere of nitrogen. The pH values of the sulphide-wool mixtures were measured at the conclusion of the experiment, using a Leeds and Northrup potentiometer, a high alkalinity glass electrode, a saturated KCl calomel electrode, and a saturated KCl bridge.

The wool was examined at regular intervals of time and classified according to the degree of retention of its fibre structure, the final observation being made after 20 hr. at 20°C. The degree of softening and disintegration of the wool has been indicated by separate symbols and plotted in Figure 9. It is

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apparent that values of pH and $\log [\text{sulphide}]$ for solutions bringing about a constant degree of wool damage, for example, that denoted by "x," fall near a straight line with unit negative slope. If the values of pH and $\log [\text{sulphide}]$ corresponding with an arbitrarily chosen degree of wool damage are plotted for digestion periods of 1, 2, 3, and 20 hr., the points approximate to a straight line in each case, the curve retaining a negative unit slope but being displaced on the coordinates with changes in time (Figure 10).

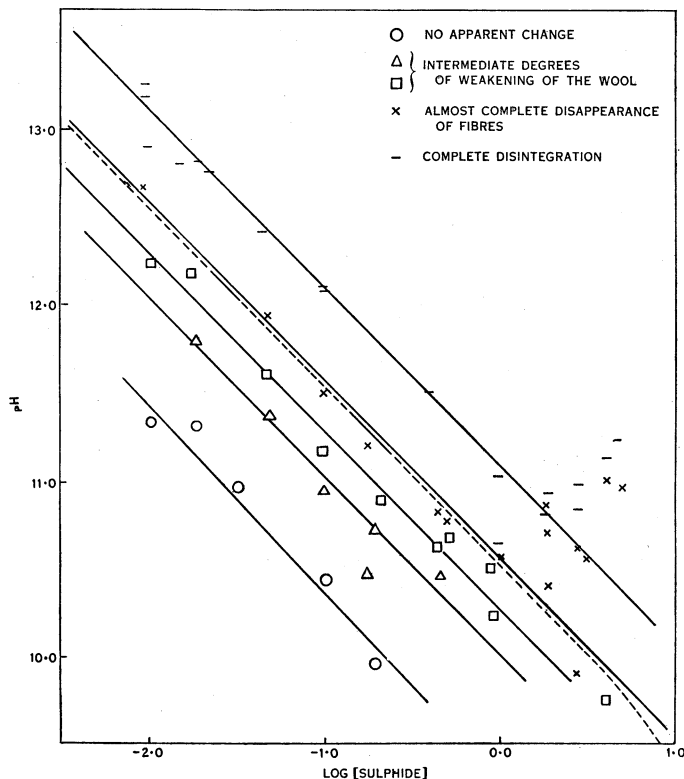


Fig. 9.—Disintegration of wool by potassium sulphide.

Since the results obtained with 3 hr. and 20 hr. digestion periods were practically identical, it is assumed that equilibrium had been reached after the latter period.

At concentrations of K_2S exceeding 1M, the results deviated from a straight line relationship (Fig. 9). This deviation from linearity was accentuated when sodium sulphide was used in place of potassium sulphide, the change then occurring at a concentration of 0.1M (Fig. 11). It was characteristic of such tests that, although the major portion of the wool was completely disintegrated, there remained a rather hard, dense residue. With sodium sulphide also, longer periods were required for the attainment of equilibrium, noticeable changes in the wool taking place for two or three days. It is probably for this reason that sodium sulphide solutions yielded more variable results than

potassium sulphide, when using a digestion period of 20 hr. Addition of sodium chloride to the sodium sulphide solutions retarded the disintegration process still further. In Figure 12, the results obtained with sodium sulphide solutions containing 2M sodium chloride are compared with the line representing almost complete disintegration of wool (\times) obtained with unsupplemented sodium sulphide solution. The digestion period was 20 hr. at 20°C. in each experiment.

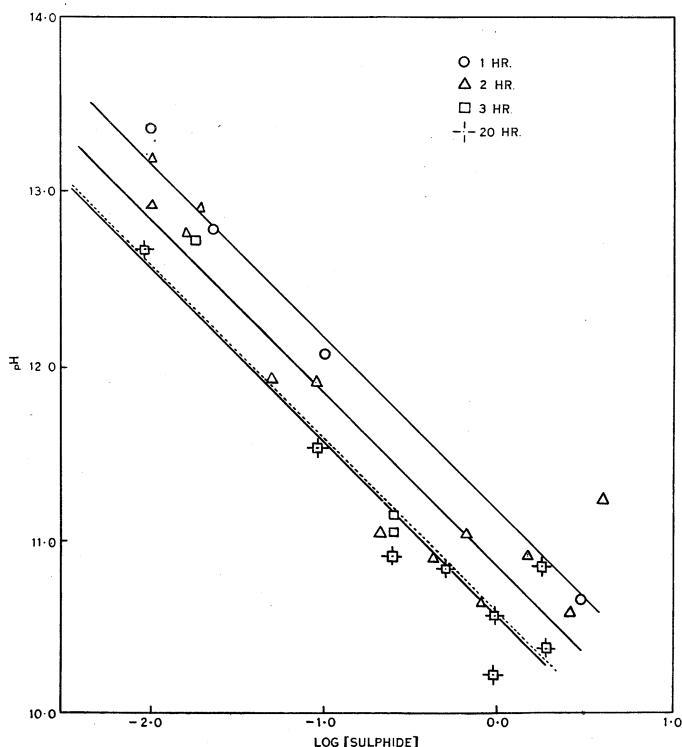


Fig. 10.—Relationship between the limiting concentrations of hydrosulphide ions and hydroxyl ions capable of reducing the strength of wool to zero.

It is evident that conditions of sulphide concentration and pH normally causing almost complete disintegration of the wool (\times) produced only partial disintegration or merely softening of the fibres (Δ or \square). However, after holding at room temperature for several days, in most instances the wool gradually softened and approached the expected equilibrium conditions. It is probable therefore that differences in the behaviour of potassium and sodium sulphide are attributable largely to slower diffusion of the more hydrated sodium ions into the wool fibre. This would have the triple effect of reducing the rate of diffusion into the wool of hydrosulphide and hydroxyl ions, with a concomitant lessening of hydration of the fibres by an osmotic effect. The latter effect would be increased by adding sodium chloride. The rate of depilation is decreased in a similar manner by high concentrations of the depilatory (Fig. 5).

Reduction of wool by sulphides involves oxidation of the sulphide to persulphide or some secondary oxidation product. The effect of persulphide on reduction of wool was determined by adding sufficient sulphur to the sulphide solutions to produce approximately equal concentrations of sulphide and persulphide. These solutions were included in an experiment of the usual type and in Figure 11 the results are compared with those given by sodium sulphide solutions. It is apparent that persulphide does not lessen the weakening of wool by sulphide but rather enhances it slightly, except at sulphide concentrations greater than 0.1M.

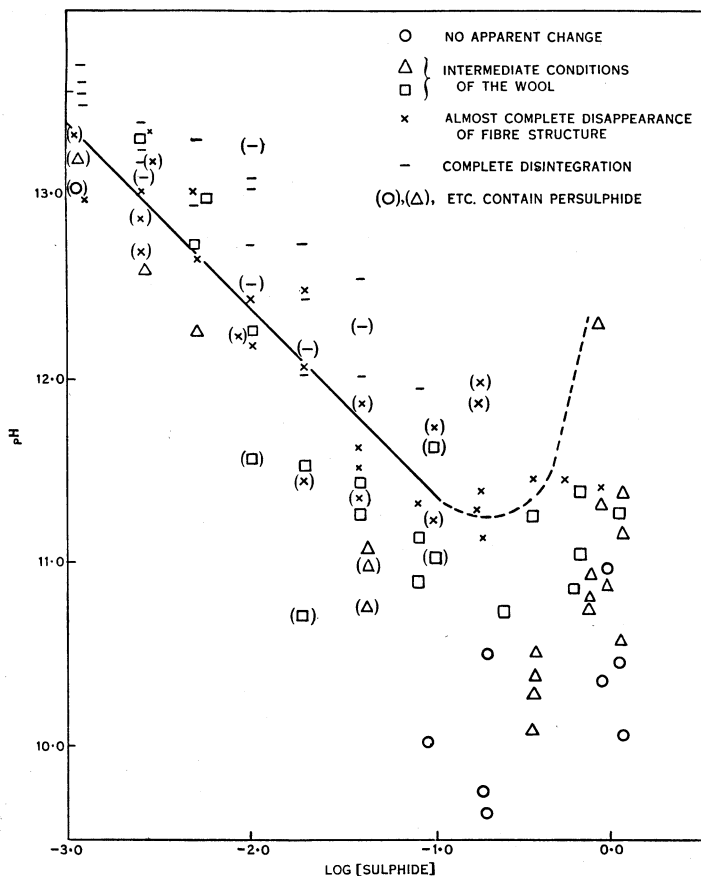


Fig. 11.—Effect of persulphide on the disintegration of wool by sodium sulphide. The curve represents the points denoted by x.

From Figure 9 it follows that, over a limited pH range, the concentration of sulphide and the pH of solutions causing a constant degree of disintegration of wool fibres may be related by the expression

$$\text{pH} = -\log [\text{sulphide}] + C, \quad \dots \dots \dots (1)$$

which, if activity coefficients are neglected, may be rewritten

$$[\text{OH}^-] [\text{sulphide}] = C_1. \quad \dots \dots \dots (2)$$

Possible explanations for this relationship fall into three main classes: (1) Higher alkalinity may increase the accessibility of disulphide bonds to sulphide; (2) Hydroxyl ions, by rupturing salt links and hydrogen bonds, may contribute directly to the weakening of wool fibres (Blackburn and Lindley 1948); (3) Hydrogen or hydroxyl ions may have a direct role in the reduction of disulphide linkages by sulphides. It is probable that more than one of the hypotheses to be examined are operative simultaneously, but since a simple relationship exists between pH and $\log [\text{sulphide}]$ over a restricted range of these values, it is almost certain that over this pH range one effect preponderates to such an extent that secondary reactions may be neglected.

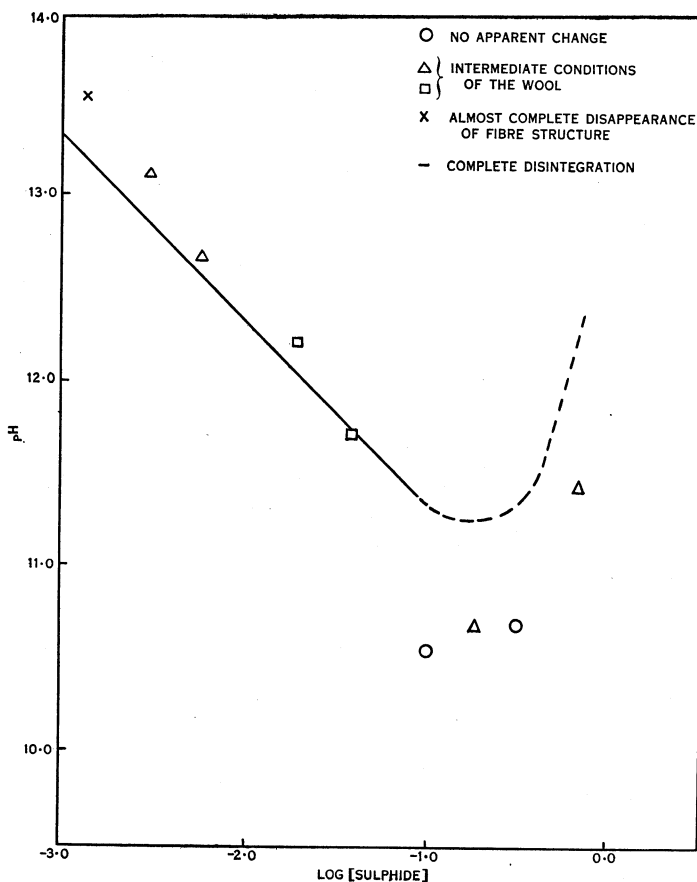


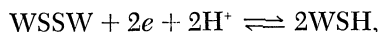
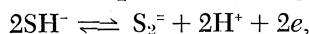
Fig. 12.—Effect of 2M sodium chloride on the disintegration of wool by sodium sulphide. The curve represents that normally joining points denoted by x.

If, as the first hypothesis suggests, the rate of diffusion of sulphide into the wool fibres were increased by increasing pH, the slope of the curve relating pH and sulphide concentration in solutions bringing about a constant degree of disintegration of wool would vary with time, and as the system approached equilibrium the slope would approach infinity. This is not substantiated by

experiment (Fig. 9). It might also be suggested that dithiol linkages are inaccessible to hydrosulphide ions until certain hydrogen bonds or salt linkages are split. However, this could not be a satisfactory explanation if a dynamic equilibrium exists between hydrogen bonds or salt linkages and the groups contributing to their formation. The ease with which such linkages are broken and reformed indicates that such an equilibrium exists (Blackburn and Lindley 1948).

The second hypothesis suggests that sulphide and hydroxyl ions act independently on the wool fibre, the former rupturing dithiol bonds, the latter breaking hydrogen bonds or salt linkages. It is, in fact, possible but highly improbable that the equations relating the decrease in strength of wool to sulphide concentration and hydroxyl ion concentration, respectively, should be such that the effects of these ions are reciprocal.

The third hypothesis suggests that pH influences the reaction between disulphide bonds of wool and sulphide. If the oxidation of sulphide and reduction of wool dithiol bonds take place according to the equations



the following electrode equations hold at equilibrium:

$$E_h = E_1^\circ - \frac{RT}{2F} \ln \frac{[\text{SH}^-]^2}{[\text{S}_2^{=}] + \frac{RT}{F} \ln [\text{H}^+], \quad \dots \dots (3)$$

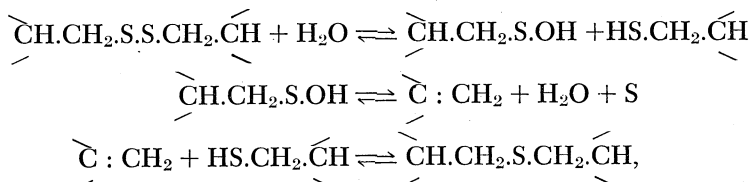
$$E_h = E_2^\circ - \frac{RT}{2F} \ln \frac{[\text{WSH}]^2}{[\text{WSSW}] + \frac{RT}{F} \ln [\text{H}^+], \quad \dots \dots (4)$$

where [WSH] and [WSSW] represent the concentrations of wool thiol and dithiol groups respectively.

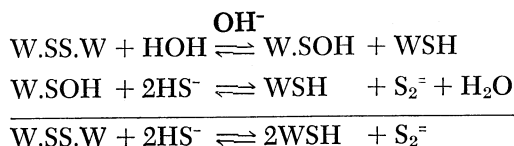
When both systems are present in equilibrium, therefore,

$$\frac{[\text{SH}^-]^2}{[\text{S}_2^{=}] + \frac{RT}{F} \ln [\text{H}^+]} = \frac{[\text{WSH}]^2}{[\text{WSSW}] + \frac{RT}{F} \ln [\text{H}^+]} \text{ antiln } \frac{2F}{RT} (E_1^\circ - E_2^\circ). \quad \dots \dots (5)$$

Cuthbertson and Phillips (1948) have shown that dithiol linkages in alkaline solution react as follows:



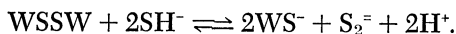
the stable lanthionine linkage being formed in this way. Bowes (1949) suggests that in strongly reducing solutions the group =CH.CH₂.SOH is reduced to =CH.CH₂.SH, thus preventing the formation of the stable lanthionine linkage. The overall reaction with sulphide as reducing agent would then be represented by



Since the final equation does not differ from that used in deriving (5), the equilibrium state would again be described by this equation. The results cannot therefore be explained by a catalytic role of OH^- .

Vago (1937) suggested that the effect of pH can be explained in terms of the decrease in Eh of sulphide solutions with increase in pH. However, the possible effect of pH on the thiol system of keratin was not considered and, unless it is postulated that one of the components of this system is involved in a secondary reaction with H or OH^- , the Eh of the wool thiol system would also decrease with increasing pH, and the equilibrium condition would again be described by (5).

The pK values of the thiol groups of reduced wool are not known. However, the pK values of the thiol groups of cysteine, glutathione, and thioglycollic acid are known to approximate to 9.0 and it seems probable therefore that the thiol groups of reduced wool should have pK values of that order. These thiol groups would then be almost completely ionized over the pH range 11.0 to 13.5 and the reactions taking place would be described by the equation



If the total concentration of sulphhydryl groups in the wool, $[\text{WSH}] + [\text{WS}^-]$, is represented by $[\text{WR}]$,

$$K_{\text{WSH}} = \frac{[\text{WS}^-][\text{H}^+]}{[\text{WSH}]}$$

$$\text{and } [\text{WR}] = [\text{WSH}] + [\text{WS}^-]$$

$$\text{Therefore } [\text{WSH}] = \frac{[\text{WR}][\text{H}^+]}{K_{\text{WSH}} + [\text{H}^+]}. \quad \dots \quad (6)$$

Eliminating WSH from (5) and (6) and converting to logarithms, we obtain

$$\begin{aligned} -\log [\text{H}^+] = & -\log [\text{SH}^-] - \log ([\text{H}^+] + K_{\text{WSH}}) + \frac{1}{2} \log \frac{[\text{WR}]^2 \text{S}_2^{=}}{\text{WSSW}} + \\ & \frac{0.4343F}{RT} (E_1^\circ - E_2^\circ). \quad \dots \quad (7) \end{aligned}$$

For values of H^+ considerably smaller than K_{WSH} , the term

$$\begin{aligned} -\log [\text{H}^+] - \text{pH}_s^* = & -\log [\text{SH}^-] + \frac{1}{2} \log \frac{[\text{S}_2^{=}] [\text{WR}]^2}{[\text{WSSW}]} + \\ & \frac{0.4343F}{RT} (E_1^\circ - E_2^\circ) + \text{p}K_{\text{WSH}}. \quad \dots \quad (8) \end{aligned}$$

Since the amount of sulphide oxidized would be equivalent to the amount of wool reduced, restriction of (8) to a constant degree of wool weakening would lead to constant values of $[\text{S}_2^{=}]$, $[\text{WSSW}]$, $[\text{WR}]$, E_1° , E_2° , and $\text{p}K_{\text{WSH}}$, so that the expression, apart from variation in activity coefficients, represents a linear relationship between pH_s and $\log [\text{HS}^-]$ with a slope of -1 .

During the disintegration of wool there will also be a free energy change due to the increased homogeneity of the system and the equilibrium state would be influenced accordingly. However, if the rupture of dithiol linkages is the major cause of wool disintegration at high pH values, it is reasonable

* Using the notation of Ogston (1947).

to assume that the decrease of free energy due to increased homogeneity of the system can be expressed as a function of $[WR]$. It would then be possible to correct for this effect by including a term $\phi ([WR])$ in (4) and (8). Since $[WR]$ is constant for a constant degree of wool disintegration $\phi ([WR])$ would also be constant.

A more rigorous treatment with the inclusion of activity coefficients in (5) and (6) gives an expression for $-\log (H^+)$ similar to (8) but with the additional term

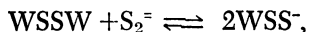
$$\frac{1}{2} \log \frac{f_{WS^-}^2 \cdot f_{H^+}^2 \cdot f_{S_2^-}}{f_{SH^-}^2 \cdot f_{WSSW}}$$

on the right hand side.

It is difficult to assess the possible influence of variation of activity coefficients on the linearity and slope of the curve. However, in considering solutions described by (8) it should be appreciated that, with decreasing ionic strength due to potassium sulphide, the ionic strength due to potassium hydroxide increases, a considerable concentration of potassium ions being built up during pH adjustment because of the buffer capacity of wool and the sulphide solution. Furthermore, the pH values obtained using the glass electrode would also be influenced by the activity coefficients of other ions in solution.

The curve obtained by assuming pK_{WSH} to be 9.0, and arbitrarily substituting 1.55 for the constant terms of (8), is represented by the broken line in Figure 9. It is seen to approximate closely to the points representing almost complete breakdown of the wool, which are denoted by \times , except at values of [sulphide] greater than 1.0M, where high salt concentrations complicate the issue.

In deriving (8), it has been assumed that the persulphide formed by oxidation of hydrosulphide ions remains completely ionized at the pH of the experiment. Although hydrogen persulphide is a much stronger acid than hydrogen sulphide, this assumption is probably unjustified in the light of what little is known of the pK values of hydrogen persulphide (Küster and Heberlein 1905). On the other hand, the results presented in Figure 11 indicate that persulphide does not decrease the disintegration of wool by sulphide and, in fact, at high concentration, it slightly increases the disintegration. It must be concluded therefore that if persulphide is a major product of the reaction, it enters into a secondary reaction, which offsets the tendency for the reverse reaction to take place. This may be due to reaction between persulphide and wool dithiol linkages.



or to further oxidation of the persulphide by wool to other polysulphides. Whatever the reason for this effect, it is apparent that small changes in $[S_2^-]$ due to partial association to HS_2^- are unlikely to affect the degree of wool disintegration significantly.

By combining (3) and (8) or by direct substitution for [WSH] in (4), an expression is obtained for the relationship between Eh of solutions and the number of residual disulphide bonds thus:

$$Eh = E^{\circ}_2 - \frac{RT}{2F} \ln \frac{[WR]^2}{[WSSW]} + \frac{RT}{F} \ln K_{WSH} \dots \dots (9)$$

This equation, like (8), is restricted in its application to a system in which $[H^+]$ is considerably less than K_{WSH} . Again it is clear that, apart from variation of activity coefficients, for a constant degree of wool weakening the Eh of the solution would be constant since WR and WSSW would be constant. Vago (1937) states that the tensile strength of horse hair keratin was reduced by sulphide solutions having reduction potentials in the range -0.3 to -0.4 V., the Eh of solutions just producing complete disintegration of the hair being constant and independent of pH. A similar range of potentials is required for the depilation of sheep skins (Fig. 7). Any hypothesis providing a satisfactory explanation of the relationship between $[OH^-]$ and [sulphide] for a constant degree of wool weakening will necessarily also provide an explanation for the constancy of Eh of such solutions as reported by Vago. This observation therefore provides experimental verification for the relationship expressed in (2). It should be pointed out, however, that, in similar experiments conducted by Vago, the slope of the straight line relating pH and sulphide was -0.7 instead of -1.0 . Since only four points are plotted in Vago's figure and in view of the difficulties of such experiments it seems probable that -1.0 is nearer the correct value, though some deviation from unit slope or a straight line relationship may occur as a result of variation of activity coefficients. The direct estimation of sulphydryl groups formed in reduced wool would probably provide more precise data.

In conclusion, it may be stated that the linear relationship between pH and the logarithm of the sulphide concentration, for solutions in equilibrium with wool that has undergone a constant degree of reductive degradation, is adequately explained by assuming the sulphydryl groups of wool to have a pK value of approximately 9.0. The fact that the Eh of such solutions is constant and independent of pH is also adequately explained on this basis.

CORRIGENDA

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Opposite page 354, Plate 1

Owing to turning of the block, Figure 1 is at bottom right; Figure 2 at bottom left; Figure 3 at top right; and Figure 4 at top left position of the plate.

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Page 491. In the caption below Figure 2: *for* 0.75M. *read* 0.075M.