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The stability of disodiumsulphosuccinated undecylenic monoethanolamide in shampoo formulations

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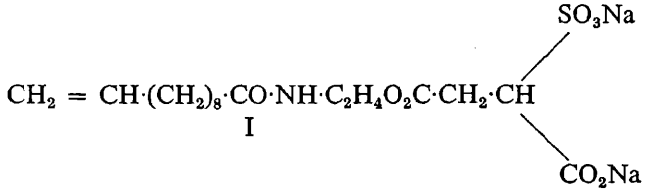
Synopsis

Disodiumsulphosuccinated undecylenic monoethanolamide (DSUM) though stable in the pH range 5·0 to 6·5, undergoes hydrolysis at pH 7 forming **undecylenic monoethanolamide.** An adaptation of an established colorimetric method based on the formation of iron (III) hydroxamates by reaction of the ester group in the molecule has been shown to offer sufficient specificity for DSUM for use in stability studies. Simultaneous screening with thin layer chromatography confirmed the specificity.

Introduction

Disodium sulphosuccinated undecylenic monoethanolamide (1) (DSUM) is used in shampoo formulations, at concentrations of about 1% w/w, as an antidandruff agent and preservative. The commercially available material is probably a mixture of related compounds but consists essentially of undecylenic monoethanolamide sulphosuccinate (I).



DSUM can be determined in shampoo formulations based on triethanolamine lauryl sulphate by a colorimetric procedure depending on the formation of iron (III) hydroxamates. The procedure employed is a modification of that described by Bergmann (2) for amides and by Goddu, LeBlanc & Wright (3) for esters. It has been found satisfactory when used as part of a production control scheme, providing a suitable 'blank' shampoo containing no DSUM is available as a comparison. The 'blank' is necessary to provide a measure of the colour obtained in the absence of DSUM due to other shampoo ingredients. When this assay procedure was applied to samples of shampoos stored for periods of several months, it appeared that the DSUM content was decreasing with time. It therefore became important to establish whether the results reflected a decrease in

DSUM content or a change in some other ingredient of the formulation. Thus the reason for the loss of DSUM, the routes of breakdown of DSUM and the specificity of the analytical method all required investigation. The results of these investigations are reported in this paper.

Routes of breakdown of DSUM—preliminary studies

The most probable route of breakdown of DSUM was thought to be by hydrolysis. Both the ester and amide functions in the molecule are susceptible to hydrolytic attack. *Fig. 1* depicts two routes of breakdown by hydrolysis. The products of alkaline hydrolysis are II and III. A white precipitate (a yellow oil if the reaction mixture is hot) is formed. The infrared spectrum of this material clearly showed the presence of an amide group (absorption bands centred at 3450, 1633 and 1550 cm^{-1}) and the presence of hydroxyl and a double bond could also be inferred, though less certainly. Bands due to sulphonate (1250 cm^{-1}) and ester groups (1740 cm^{-1}) present in the original material were missing. This information is consistent with structure II, undecylenic acid monoethanolamide. It then follows that the other product is III, trisodium sulphosuccinate which is water soluble and was not isolated from the reaction mixture. When II is treated with alkaline hydroxyammonium sulphate and subsequently with iron (III) chloride solution as in the colorimetric assay procedure (see below), no colour is produced. It is clear, then, that the ester function in DSUM is alone responsible for the chromogenic response in the assay procedure, the amide function playing no part in colour formation since the conditions are not sufficiently vigorous (2), (4). Acid hydrolysis leads to a loss of surface activity in dilute solutions (0·1% w/v) as measured by a biphasic cetrimide titration in which an indicator (methylene blue) is distributed between chloroform and dilute sulphuric acid solution to give an equal depth of colour at the end point. This loss of surface activity amounted to 50% at pH 2·65 in 24 h, and was accompanied by a loss of ester function, measured by the colorimetric procedure (see below) of less than 10%. This shows that while the ester function is not readily attacked under cold acidic conditions some other part of the molecule does undergo change.

Experimental

COLORIMETRIC DETERMINATION OF DSUM IN SHAMPOO

Reagents

All reagents were of analytical reagent grade where possible.

M Hydroxyammonium sulphate

3·5M Sodium hydroxide solution

3·5M Hydrochloric acid

Iron (III) chloride stock solution

A 0·444M solution in 0·1M hydrochloric acid.

Iron (III) chloride reagent

Dilute 5·0 ml Iron (III) chloride stock solution, filtered if necessary, and 1·0 ml 3·5M hydrochloric acid to 25 ml with distilled water.

Alkaline hydroxylamine Reagent

Mix equal volumes of M hydroxylamine sulphate solution and 3·5M Sodium hydroxide. Prepare a fresh solution daily.

Procedure

Dilute about 5 g of shampoo, (W_{sa}g,) weighed to ± 0·02 g to 50 ml in a standard flask. To a 4·0 ml aliquot in a 25 ml standard flask add 5 ml alkaline hydroxylamine reagent, swirl gently and stand at room temperature for exactly 5 min. (Use a stop watch and time the 5 min-period from the start of the addition of reagent.) Then add 5·0 ml hydrochloric acid, swirl and immediately add 1·0 ml iron (III) chloride reagent. Swirl to mix and dilute to the mark with 95% ethanol. Mix well, allow the gas evolution to subside and transfer to a 40 mm cell. Measure the absorbance (A_{sa}) of the solution within 3 min at 530 nm using water as the reference taking care to ensure there are no bubbles in the light path. Using a sample (W_{blk} of the base shampoo containing the same ingredients as the sample but lacking the DSUM carry out the same determination to obtain a blank result (A_{blk}).

Calibrations

Primary calibration with DSUM. This calibration is only required initially.

Prepare solutions of DSUM in distilled water to cover the concentration range 0·25 to 1·25 mg ml^{-1}. Take 4 ml aliquots and apply the colorimetric procedure described above. Plot a graph of the net absorbance readings *vs* DSUM (mg) taken. The plot should be a straight line passing through the origin. Calculate the slope (F) of the line in terms of DSUM (mg) per absorbance unit.

Secondary calibration with ethyl acetate. This calibration should be performed simultaneously with the primary calibration and on each occasion that DSUM is determined.

Dissolve 1 g ethyl acetate (W_{std}) accurately weighed, in 20 ml ethanol in a 50 ml standard flask. Dilute to the mark with ethanol. Mix well and transfer 2·0 ml to a 250 ml standard flask containing 150 ml distilled water and dilute to the mark with distilled water. Mix well and transfer 4·0 ml to a 25 ml standard flask and continue as in procedure from 'Add 5 ml alkaline hydroxylamine reagent . . .' measuring the absorbance (A_{std}) of the resulting solution in a 40 mm cell at 530 nm.

Calculation

DSUM in sample = $50/4\ (A_{sa} - A^*_{blk})\ F'$mg

Where A_{sa} = absorbance of sample solution at 530 nm

A^*_{blk} = absorbance of shampoo blank solution at 530 nm

corrected for weight difference = $\frac{W_{sa}}{W_{blk}} \times A_{blk}$

F' = Calibration factor which is calculated by allowing for changes in sensitivity of the method due to reagent differences, temperature differences etc. using the secondary calibration results:—

$$F' = F \times \frac{W_{std_1}}{A_{std_1}} \times \frac{A_{std_2}}{W_{std_2}}$$

in which the subscripts 1 and 2 refer to secondary calibrations performed at the same time as primary calibration and determination respectively.

THIN LAYER CHROMATOGRAPHIC EXAMINATION OF SHAMPOO

Reagents

TLC development solvent: 15 ml 1,4-dioxan, 75 ml chloroform and 15 ml carbon tetrachloride. Prepare a fresh mixture daily.

Hydrolysis products of DSUM

Alkaline hydrolysis product

Weigh 10 g DSUM. Add 100 ml M sodium hydroxide solution and reflux for 30 min. Cool and filter. Wash the precipitate with distilled water until free from alkali. Recrystallize from aqueous acetone. The yield is about 3 g.

Acidic hydrolysis product

Weigh 10 g DSUM, add 100 ml 3M hydrochloric acid and reflux for 1 h. Extract the oil or precipitate with diethyl ether. Dry by the addition of anhydrous sodium sulphate, filter and evaporate to dryness. The yield is about 2·5 g.

Precoated silica gel 60 TLC plates (E. Merck) 10 × 20 cm.

Ethanolic sulphuric acid solution

Add cautiously 10 ml concentrated sulphuric acid to 80 ml ethanol (74 o.p. spirit). Cool and dilute to 100 ml with ethanol.

Procedure

Extract 4·0 g of the sample with three separate 10 ml portions of diethyl ether. Combine the ether extracts and wash once with 20 ml distilled water. Discard the aqueous wash and dry the ether extract over anhydrous sodium sulphate. Filter, evaporate on a water bath to a small volume, transfer the residue to a 5 ml standard flask using diethyl ether, and dilute to the mark. Prepare 0·4% w/v solution of the alkaline hydrolysis product and a 0·33% w/v solution of the acid hydrolysis product. These solutions, when 5 µl is spotted directly on to the TLC plate, are equivalent to 100% degradation of the DSUM. Dilute these standard solutions to provide solutions corresponding to 25%, 50% and 75% degradation. Apply 5 µl portions of the sample extract and these diluted solutions to the plate. Develop the plate to about 15 cm above the origin line, and dry in an oven at 100°C. Spray with ethanolic sulphuric acid. Dry at 140°C for 15 min. View the plate under U.V. illumination (365nm), and compare the intensities of the standard spots with those from the sample to estimate extent of degradation. Allow for the apparent degradation simultaneously detected in an extract from a freshly prepared aqueous 1% solution of DSUM.

Results and discussion

Assay procedure

The colorimetric assay depends upon measurement of an unstable colour. All assays were therefore performed in duplicate working rapidly to ensure that absorbance measurements were complete within 3 min of the addition of the iron (III) chloride reagent. The conditions described above have been found to give optimum colour stability. The intensity of the colour produced has been found to vary with the batch of prepared reagent used. For this reason ethyl acetate was used as a convenient standard substance to monitor the sensitivity given by a batch of reagent. This avoided the use of DSUM as a

working standard which would not have been satisfactory since its long term stability was unknown.

Results obtained with this procedure over several months have shown that in routine use the 95% confidence limits (2 σ) are $\pm$ 0·05% w/w DSUM at a nominal 1% w/w concentration.

From the results of the preliminary studies it was apparent that the colorimetric assay procedure is specific for DSUM in the presence of its decomposition products formed by alkaline hydrolysis. If acidic hydrolysis occurs, the method may not produce valid results as the decomposition may proceed only as far as the formation of (IV) and (V), (*Fig. 1*), the latter containing an ester group which would respond to the colorimetric assay in a similar way to DSUM. This aspect of method specificity was investigated in conjunction with a storage test using TLC to detect degradation products.

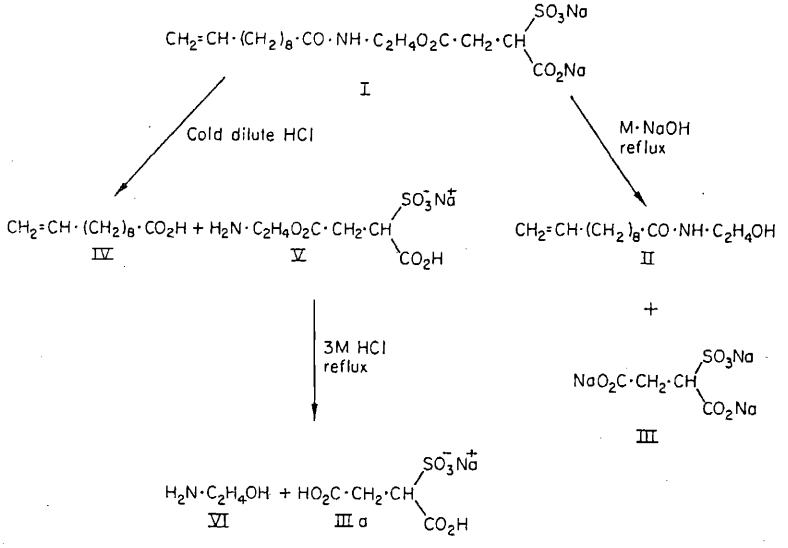


Figure 1. Routes of breakdown of DSUM by hydrolysis.

Storage test of shampoos

The second part of the investigation took the form of a storage test during which the colorimetric assay was used in conjunction with TLC screening. The stability of DSUM (1·0% w/w) in citrate buffered shampoo containing 17% triethanolamine lauryl sulphate was monitored as a function of apparent pH and of temperature. Control samples containing no DSUM were also monitored to provide 'blank' values. The DSUM assay results are presented in diagrammatic form in *Fig. 2* (*a*) and (*b*) and show clearly the dependance of stability on apparent pH. The loss of DSUM follows a first order rate equation. The concentration axes in *Fig. 2* (*a*) and (*b*) are logarithmic and the plots are therefore linear within the limits imposed by the assay procedure. At an apparent pH of 7 DSUM is rather unstable, 25% decomposing in 12 weeks at 20°, while at 37° 58% was lost over the same period. Confirmation of the validity of these results was obtained from the TLC screening test. A typical chromatogram is shown schematically in *Fig. 3*. The separated components are seen as coloured fluorescent zones. In addition to unidentified spots at R_F zero, 0·4–0·6 and 0·95 a clearly separated spot, having a pink

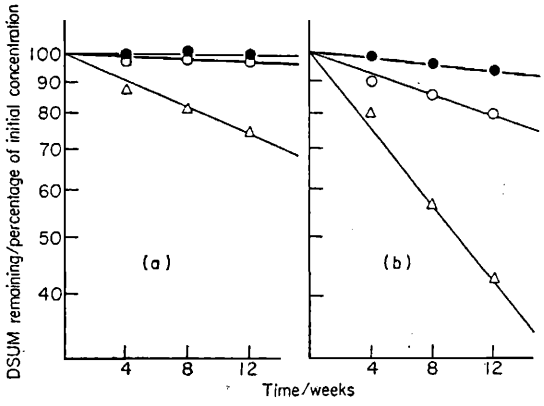


Figure 2. The loss of DSUM from shampoos at various pH values (a) at 20°C, (b) at 37°C. Key: ● pH 5·0; ○ pH 6·5; △pH 7·0.

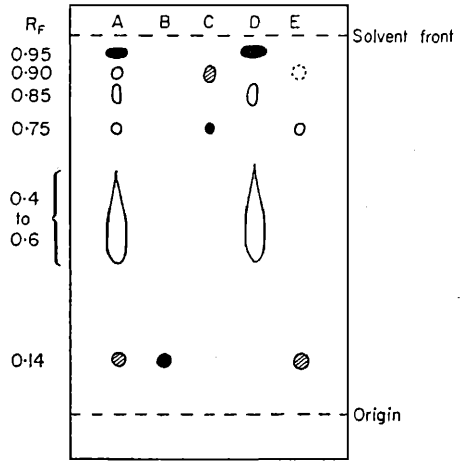


Figure 3. Schematic representation of developed TLC plate viewed under a UV lamp (365 nm). The extent of shading of the spots indicates approximate visual intensity. (A) Extract from shampoo containing DSUM and its breakdown products. (B) Solution of II—product of alkaline hydrolysis. (C) Solution of IV—product of acidic hydrolysis. (D) Extract from blank shampoo. (E) Extract from aqueous DSUM solution.

fluorescence, was seen at R_F 0·14. This corresponds in colour and mobility to II, undecylenic monoethanolamide, prepared from DSUM as described above. Trace amounts of the products of acidic hydrolysis were found in all samples including the original DSUM raw material, but only II was seen to increase in concentration on storage. Some approximate results obtained by TLC spot intensity comparisons as described above are shown in *Table I*.

From the chromatograms it was immediately evident that the shampoos at an apparent pH of 7 and stored at 37°C contained much more II than those at lower apparent pH. Furthermore the estimated quantities of II (*Table I*) corresponded quite closely with

Table I. Estimates of breakdown of DSUM from visual spot comparisons on TLC plates. Loss of DSUM (% of initial) by formation of II

	Period of storage at 37°	
pH of shampoo	8 *weeks*	12 *weeks*
5·0	$<<25$	$<<25$
6·0	$<<25$	$<<25$
7·0	~ 50	> 50

the loss of ester function as determined by colorimetry (*Fig. 2(b)*). These two observations are consistent with a hydrolysis reaction producing II and free sulphosuccinate, III, as shown in *Fig. 1*. At lower pH values this reaction does not occur and no other degradation products were observed to be produced.

It is interesting to note that the product of acidic hydrolysis, IV, was resolved into two separate components by TLC. No further work was done to identify the two components, but the hydrolysis product isolated could well have been a mixture of homologues.

Conclusions

The colorimetric assay procedure was shown to offer adequate specificity for use in stability studies of DSUM. The results of the storage test show that DSUM is sufficiently stable in the pH range 5 to 6·5 in shampoo formulations for commercial exploitation, but that at pH 7 and above significant loss occurs by hydrolysis with the formation of undecylenic monoethanolamide. The pH of such formulations should be carefully controlled to minimise the loss of DSUM. Loss of DSUM by formation of undecylenic monoalkanolamides may not however be entirely undesirable since these compounds are reported to have antimicrobial activity (5).

References

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