*J. Soc. Cosmetic. Chem.*, **28**, 521–532 (September 1977)

# The effect of detergents on swelling of stratum corneum

GERALD J. PUTTERMAN,* NANCY F. WOLEJSZA, MARIA A. WOLFRAM, and KARL LADEN † *Gillette Research Institute, 1413 Research Boulevard, Rockville, MD 20850.*

*Received October 6, 1976*

**Synopsis**

Several surfactants were tested for their ability to produce in-plane SWELLING (increase surface area) of squares of GUINEA PIG STRATUM CORNEUM. Highest levels of swelling were observed with the anionic surfactants sodium laurate and sodium lauryl sulfate, while little or no swelling was observed with the few cationic and nonionic surfactants examined. Although swelling in laurate was shown to be reversible, work index measurements revealed an irreversible weakening of the tissues. To gain insight into the mechanism of swelling the effects of protein denaturants and delipidizing agents were also evaluated. We conclude that protein denaturants, *per se*, do not cause stratum corneum swelling, but that swelling is due to a reversible conformation change resulting from cooperative binding of the detergent. Stratum corneum swelling could be of value for studying detergent-skin interactions and for predicting detergent penetration of skin and possible subsequent skin irritancy.

## INTRODUCTION

Among the properties of skin which have been shown to be altered by detergent treatment are its permeability, extractability of amino acids and Folin-Ciocalteu positive material (protein) and liberation of reactive sulfhydryl groups (1). In terms of dimensional changes Choman (2) observed increases in the thickness of epidermis-free calf skin and human abdominal skin which were produced by treatment with sodium alkyl sulfates of different chain lengths and concluded that sodium lauryl sulfate produced the greatest increase. In order for swelling to occur a concentration near or above the critical micelle concentration (CMC) of each alkyl sulfate was required. In their ultrastructural study of the action of 1 per cent sodium lauryl sulfate on rat skin Tovell and coworkers (3) noted a marked thickening of the epidermis and stratum corneum resulting from treatment with the detergent.

Scheuplein and Ross (4) soaked stratum corneum in 5 per cent sodium laurate for 24 h and observed a visible expansion in the plane of the tissue. These authors also noted that Von Götte (5) had previously observed an expansion of isolated epidermis after

*Present Address: Frederick's Cancer Research Center, P.O. Box B, Frederick, MD 21701.
†Present Address: Carter Products, Division of Carter-Wallace, Inc., Half Acre Road, Cranbury, NJ 08512.

treatment with anionic detergents. Since the measurement of surface area would be a convenient means of evaluating stratum corneum-detergent interactions, we have utilized the method to confirm the results for sodium laurate (4) and examine other detergents. To gain an insight into the mechanism of this swelling phenomenon we have also determined the effect of other agents known to interact with protein or lipid on stratum corneum surface area and have evaluated the effect of sodium laurate on a rheological property of stratum corneum.

## MATERIALS

Stratum corneum was obtained from Hartley guinea pigs. Excised, epilated* skin was exposed to ammonia vapor (6). The sheets were air dried and stored under ambient conditions.

Analytical grade reagents were used without further purification. These included urea, guanidine hydrochloride, dimethylsulfoxide, formic acid, ammonium thioglycollate, dithiothreitol, lithium bromide, N-methyl-2-pyrrolidone, chloroform, methanol, and ethyl ether. The sources of surfactants used in this work are listed in Table I.

## METHODS

### IN-PLANE SWELLING

Squares of stratum corneum 20 × 20 mm were soaked in water for 1 h. The squares were lifted out of the water on plastic screens and their dimensions were determined with a ruler calibrated in millimeters. The squares were then immersed in the appropriate solution for 16 h after which they were removed on plastic screening, and their dimensions were again determined. Swelling is expressed as the percent change in area after exposure to the second solution as compared to the first solvent, water.

### MECHANICAL TESTING OF STRATUM CORNEUM

Strips of stratum corneum, prepared according to procedures previously described (7), were immersed in water for 4 h and stretched 5 per cent of their original length. After relaxation they were exposed to a test material for 1 h, followed by transfer to water and a second stretching after 4 h. The results are expressed in terms of a work index (7) where:

$$\text{work index} = \frac{\text{work required to stretch strip after treatment}}{\text{work required to stretch strip before treatment}}$$

### STRATUM CORNEUM MODIFICATION

1. *Oxidation:* Performic acid was prepared by combining 9 parts of concentrated formic acid (98 per cent) with 1 part hydrogen peroxide (30 per cent) and letting the mixture stand for 2 h at room temperature. The performic acid was diluted 1:5 with water just before using on stratum corneum in a bath ratio of 100:1 at 15–20°C for 30 min. The stratum corneum was rinsed well in deionized water. An 88 per cent oxida-

*Zip Wax®, Jean Jordeau Inc., New York.

Table I
Surfactants Studied

| Name | Concentration | Manufacturer | CMC[a] |
|---|---|---|---|
| Sodium laurate | .05 *M* | Eastman Organic Chemicals, Rochester, NY | $2.4 \times 10^{-2}$ *M* |
| Sodium lauryl sulfate | .05 *M* | Fisher Scientific Corp., Fairlawn, NJ | $8.1 \times 10^{-3}$ *M* |
| Ammonium lauryl sulfate | .05 *M* | Continental Chemical Co., Clifton, NJ | $6.0 \times 10^{-3}$ *M* |
| Triethanolammonium lauryl sulfate | .05 *M* | Alcolac Inc., Baltimore, MD | $4.0 \times 10^{-3}$ *M* |
| Sodium dodecylbenzene sulfonate | .05 *M* | Pfaltz and Bauer, Inc., Flushing, NY | $1.2 \times 10^{-3}$ *M* |
| Sodium myristal ether sulfate | .05 *M* | Standapol ES-40 Conc.—Henkel, Inc., Teaneck, NJ | $1.4 \times 10^{-3}$ *M* |
| Sodium lauryl ether sulfate | .05 *M* | Sipon ES—Alcolac Inc., Baltimore, MD | $4.8 \times 10^{-3}$ *M* |
| Sodium alkyl sulfonate | .05 *M* | Ultrawet K—ARCO Chemical Co., Philadelphia, PA | $9.0 \times 10^{-3}$ *M* |
| Sodium oleate | .05 *M* | Fisher Scientific Corp., Fairlawn, NJ | $2.6 \times 10^{-3}$ *M* |
| Sodium hexanoate | .05 *M* | Eastman Organic Chemicals, Rochester, NY | 1.6 *M* |
| Lauryl isoquinolinium bromide (Q-75) | .05 *M* | Q-75—Onyx Chemical Co., Jersey City, NJ purified from Isothan | $4.8 \times 10^{-3}$ *M* |
| Stearyl dimethyl benzyl ammonium chloride | 20 per cent | Triton X-400—Rohm & Haas Co., Philadelphia, PA | $8.5 \times 10^{-6}$ *M* |
| Brij 35, pH 4.8 | .05 *M* | Atlas Chemical Co., Wilmington, DE | $6.0 \times 10^{-5}$ *M* |
| Octylphenoxy polyethoxy ethanol | 1 per cent | Triton X-100—Rohm & Haas Co., Philadelphia, PA | $2 \times 10^{-3}$ per cent |

[a] Values obtained from (17).

Table II
Effect of Anionic Surfactants on Stratum Corneum Swelling

| Surfactant | Conditions | Per cent Swelling | Standard Deviation | n[a] |
|---|---|---|---|---|
| Sodium laurate | .05 *M*, pH 9.8 | 16.7 | ±9.7 | 37 |
| Sodium lauryl sulfate | .05 *M*, pH 10 | 13.1 | ±3.5 | 97 |
| Ammonium lauryl sulfate | .05 *M* | 9.1 | ±2.9 | 4 |
| Triethanolammonium lauryl sulfate | .05 *M* | 12.2 | ±2.3 | 4 |
| Sodium dodecylbenzene sulfonate | .05 *M* | 11.9 | ±10.8 | 4 |
| Sodium myristyl ether sulfate | .05 *M* | −0.1 | ±1.4 | 7 |
| Sodium lauryl ether sulfate | .05 *M* | 6.6 | ±2.9 | 4 |
| Sodium lauryl triether sulfate | .05 *M* | 3.9 | ±3.8 | 4 |
| Sodium oleate | .05 *M* | 15.1 | ±7.9 | 6 |
| Sodium hexanoate | .05 *M* | 0.0 | ±0.0 | 4 |

[a]Number of measurements.

tive fission of disulfide bonds was obtained as determined by amino acid analyses of acid hydrolysed samples.

2. *Reduction and Alkylation:* For the reduction of stratum corneum 0.02 *M* dithiothreitol was used at 200:1 bath ratio for 1 h at 39°C. After reduction stratum corneum was alkylated with 2.5 per cent acrylonitrile in 1 per cent borate buffer (pH 9.1) for 30 min at 35°C in a 50:1 bath ratio. Alternatively, the stratum corneum was alkylated with 2 per cent iodoacetate in borate buffer (pH 8) for 2 h at 35°C in a 100:1 bath ratio. The reaction flask was evacuated during the reaction.

### DELIPIDIZATION

Delipidization was obtained with $CHCl_3$:MeOH(2:1) for 90 min at room temperature followed by 5 min extraction with water.

## RESULTS

Several anionic surfactants at concentrations above their CMC (Table I) were tested for their ability to produce in-plane swelling (increase surface area) of guinea pig stratum corneum squares. The results given in Table II confirm the observation of Scheuplein and Ross (4) for sodium laurate and indicate that sodium lauryl sulfate produces the same effect. The increase in surface area obtained with sodium laurate is illustrated in Fig. 1. The sodium cation is not necessary for the effect since similar levels of swelling were found when ammonium and triethanolammonium lauryl sulfate were used. Sodium dodecyl benzene sulfonate and sodium oleate also gave high levels of in-plane swelling, while sodium hexanoate gave no measurable swelling. This absence of swelling with hexanoate could be a direct effect of using a short chain molecule (i.e., a short chain molecule might not interact with stratum corneum as readily as the longer laurate) or the effect might be indirect in that the concentration of hexanoate employed, although equal to that of the other anionic surfactants, was below its CMC (Table I). (In this regard, 0.005 *M* sodium lauryl sulfate produced no measurable swelling

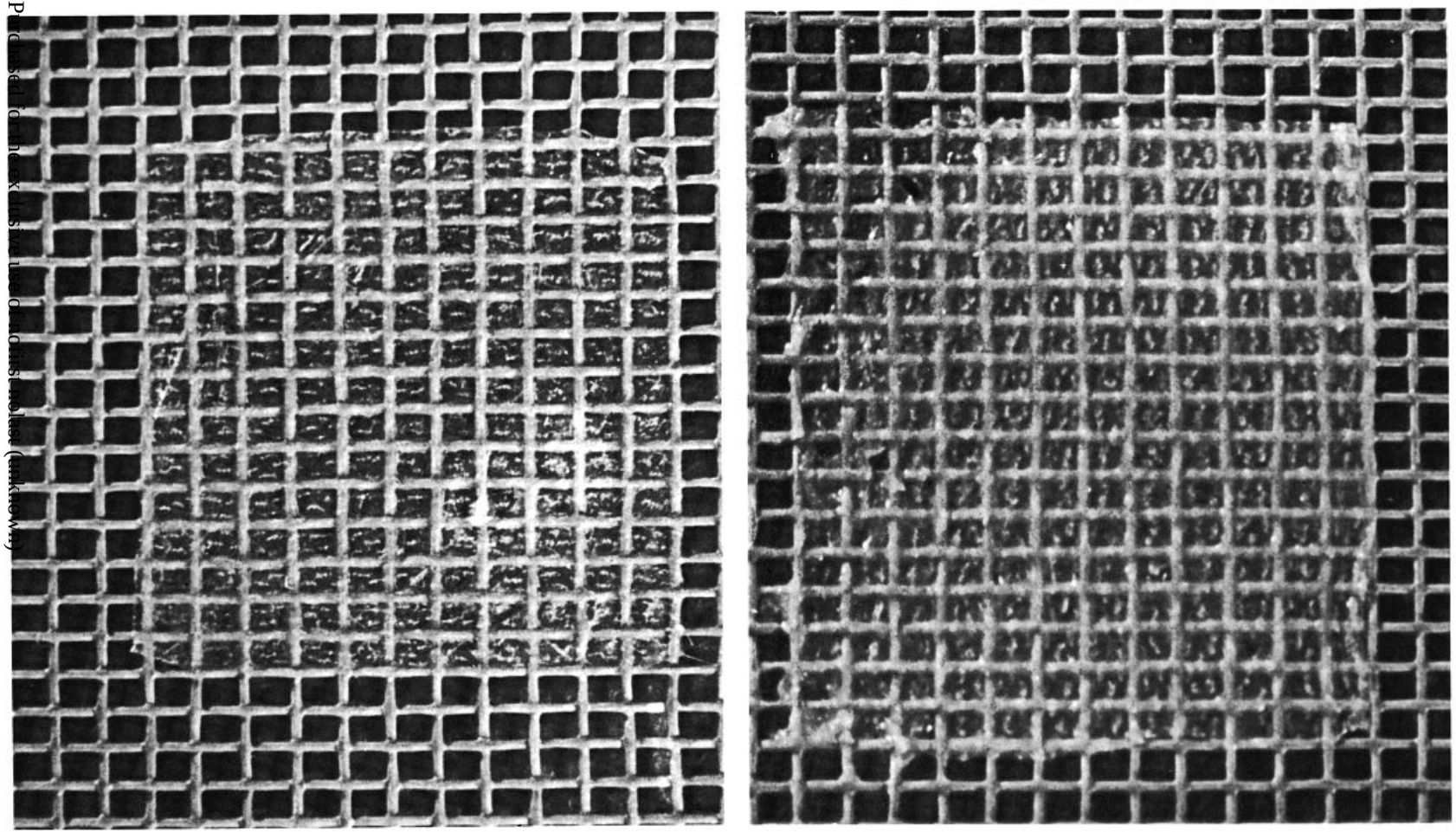


**Figure 1.** Illustration of in-plane swelling of stratum corneum in sodium laurate: a) following initial water soak, b) the same piece of stratum corneum following sodium laurate soak

Table III
Effect of Cationic and Nonionic Surfactants on Stratum Corneum Swelling

| Surfactant | Conditions | Per cent Swelling | Standard Deviation | n[a] |
|---|---|---|---|---|
| Laurylisoquinolinium bromide (Isothan Q-75) | .05 *M* | 3.9 | ±3.8 | 4 |
| Stearyldimethylbenzylammonium chloride (Triton X-400) | 20 percent | 1.6 | ±0.7 | 4 |
| Polyoxyethylene (23) lauryl ether (Brij 35) | .05 *M*, pH 4.8 | 3.4 | ±3.4 | 4 |
| Octylphenoxypolyethoxyethanol (Triton X-100) | 0.1 percent | 3.1 | ±0.9 | 4 |

[a]Number of measurements.

Table IV
Effects of Protein Denaturants and Lipid Solvents on Stratum Corneum Swelling

| Reagent | Type[a] | Per cent Swelling | Standard Deviation | n[b] |
|---|---|---|---|---|
| 8 *M* urea | D | 0 | ±0.0 | 4 |
| 5 *M* guanidine-HCl | D | 4.1 | ±1.6 | 4 |
| 0.1 *M* ammonium thioglycollate | D | 0 | ±0.0 | 4 |
| 0.1 *M* DTT | D | 0 | ±0.0 | 4 |
| 50 per cent LiBr | D | 0 | ±0.0 | 4 |
| 80 per cent dimethylsulfoxide in water | L | 0 | ±0.0 | 4 |
| 80 per cent N-methylpyrrolidone in water | L | 0 | ±0.0 | 4 |
| Chloroform: methanol (2:1) | L | 0.6 | ±1.0 | 10 |
| ethyl ether | L | −4.1 | ±1.5 | 6 |
| 0.3 per cent performic acid, 15°C | D | 26.3 | ±12.9 | 10 |

[a]D is a protein denaturant; L is a lipid solvent.
[b]Number of measurements.

Table V
Effect of Pretreatment on Sodium Lauryl Sulfate-Induced Swelling of Stratum Corneum

| Pretreatment[a] | Per cent Swelling in[b] 0.05M NaLS (pH 6) | Standard Deviation | n[b] |
|---|---|---|---|
| $H_2O$ | 13.1 | ±3.5 | 97 |
| $CHCl_3$:MeOH delipidized | 24.3 | ±6.2 | 8 |
| DMSO | 36.8 | ±2.8 | 4 |
| DTT reduced | 14.2 | ±0.1 | 4 |
| DTT-acrylonitrile alkylated | 16.0 | ±6.3 | 10 |
| DTT-iodoacetate alkylated | 23.4 | ±5.2 | 8 |
| Performic acid oxidized | −1.7[c] | ±3.2 | 4 |

[a]Abbreviations: DMSO-dimethyl sulfoxide; DTT-dithiothreitol.
[b]Number of measurements.
[c]Performic acid oxidation induced a swelling of 26 per cent, which was not reversible in water. No additional swelling occurred upon exposure to NaLS.

while readily detectable swelling was generated in 0.01 *M* sodium lauryl sulfate.) That a hydrophobic chain in the surfactant favors stratum corneum swelling is indicated by a comparison of results obtained for sodium lauryl sulfate and sodium lauryl ether sulfate (1 mol of ethylene oxide). Sodium lauryl ether sulfate with 3 mol of ethylene oxide

yielded an even lower level of stratum corneum swelling. Sodium myristyl ether sulfate (1 mol of ethylene oxide) produced no swelling indicating that the combination of increasing polarity and a $C_{14}$ alkyl group instead of $C_{12}$ eliminated swelling.

To determine whether the in-plane swelling of stratum corneum was produced only by anionic surfactants, two cationic surfactants (Triton X-400) and Isothan Q-75) and two nonionic surfactants (Brij 35 and Triton X-100) were evaluated. The results shown in Table III indicate that little swelling is obtained with these surfactants compared to levels found for long chain anionic surfactants.

Since sodium lauryl sulfate has been used to denature proteins,* other agents which denature proteins (probably by means of a different mechanism) were evaluated for their ability to swell stratum corneum (Table IV). Reagents which are strong hydrogen bond formers (5 *M* guanidine hydrochloride, 8 *M* urea), reducing agents (ammonium thioglycollate, dithiothreitol), and 50 per cent lithium bromide produced little or no stratum corneum swelling. Performic acid oxidation run under mild conditions (0.3 per cent performic acid, 15°C, 30 min) produced extensive swelling which probably reflects its greater efficiency (compared to reducing agents) in cleaving disulfide bonds as well as the effect of introducing hydrophilic negative sites into the keratin. Agents capable of removing lipid (80 per cent dimethyl sulfoxide,† 80 per cent N-methyl pyrrolidone in water,† and chloroform:methanol (2:1 by volume) produced no swelling.

Not only was stratum corneum swelling measured in the protein denaturants cited above but stratum corneum soaked in some of these agents was subsequently exposed to the known swelling agent sodium lauryl sulfate. The results are summarized in Table V. Delipidizing agents and reducing agents appear to enhance sodium lauryl sulfate induced swelling while performic acid oxidation does not. Since per cent swelling in sodium lauryl sulfate is calculated by comparing the stratum corneum dimensions in the surfactant to its dimensions in water following the pretreatment, it would appear that the tissues have reached their maximum capacity of swelling in performic acid.

Since the greatest increases in swelling were produced by sodium laurate and sodium lauryl sulfate, these surfactants were further examined. If surfactant-induced swelling was specifically due to the extraction of proteins or lipids from the stratum corneum, one would expect that the swelling effect would be irreversible. However, reversibility of the effect was demonstrated by alternately swelling stratum corneum in sodium laurate and returning it to its original size in water (Table VI).

Although the swelling in sodium laurate appeared to be reversible, it seemed reasonable to examine the effect of laurate on other properties of the stratum corneum. Thus, stratum corneum strips were first stretched on the Instron extensometer approximately 5 per cent in water, and after relaxation were exposed to either water, 0.05 *M* sodium laurate at pH 9.8 or 0.05 *M* sodium acetate at pH 9.8 for 1 h followed by transfer to water and a second stretching. The work index (defined in the Methods Section of this paper) would be 1.0 if the treatment had no effect and $<1$ if the treat-

*Sodium lauryl sulfate has been used as a denaturing agent in the determination of protein molecular weights (8).

†These aprotic solvents are also capable of forming hydrogen bonds and have often been classified as protein denaturants.

Table VI
Sodium Laurate (NaL) Induced Swelling of Stratum Corneum—Reversibility Studies[a,b]

| | Per Cent Swelling in 1 Per Cent NaL | Standard Deviation | Per Cent Swelling in $H_2O$[c] | Standard Deviation | Per Cent Swelling in 1 Per Cent NaL | Standard Deviation | Per Cent Swelling in $H_2O$[c] | Standard Deviation |
|---|---|---|---|---|---|---|---|---|
| Animal I | 24.6 | ± 4.9 | −1.4 | ± 2.4 | 22.8 | ±3.4 | 0.6 | ±2.7 |
| Animal II | 25.3 | ±12.5 | 0.0 | ±0.0 | 20.1 | ±7.6 | −6.4 | ±2.7 |

[a] 0.05 *M* sodium laurate, pH 9.8.
[b] Each value represents a measurement on 3 pieces of stratum corneum.
[c] Recovery time in water is 4 h.

Table VII
Effect of Sodium Laurate (NaL) on Work Index of Stratum Corneum

| Treatment | Work Index | Standard Deviation | n[a] |
|---|---|---|---|
| Water | 0.98 | ±0.07 | 7 |
| 1 per cent sodium acetate, pH 9.8, 60 min | 0.88 | ±0.07 | 5 |
| 1 per cent NaL, pH 9.8, 60 min | 0.50 | ±0.17 | 7 |

[a]Number of measurements.

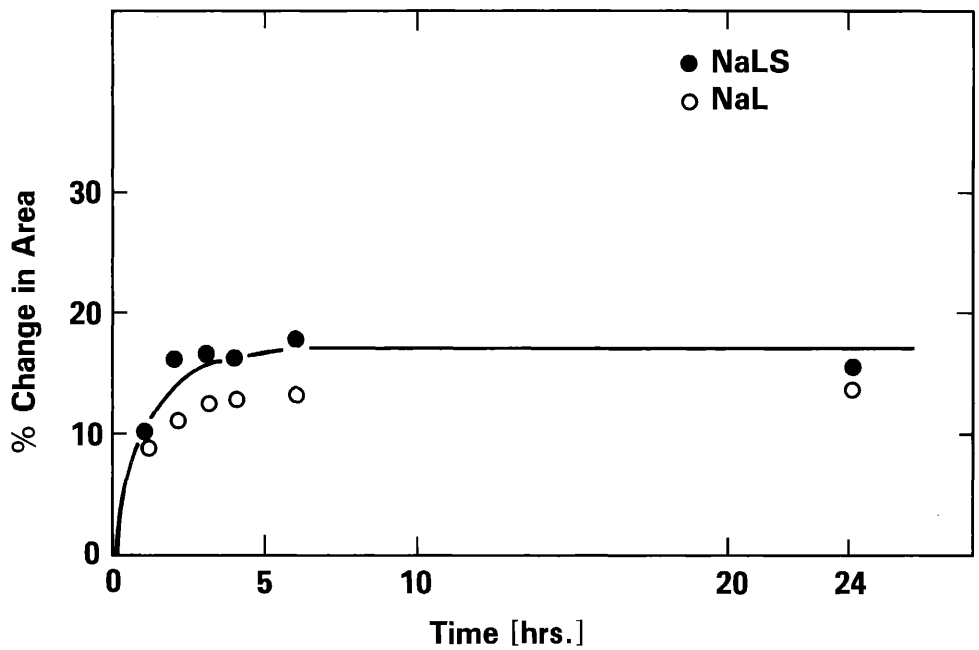


Figure 2. Rate of in-plane swelling in sodium lauryl sulfate (NaLS) and sodium laurate (NaL)

ment produced weakened stratum corneum. The data in Table VII indicate that water and sodium acetate had little effect on the strength of the tissue while sodium laurate produced a notable weakening. Rate studies of stratum corneum swelling in sodium laurate and sodium lauryl sulfate (Fig. 2) justified the brief exposure to surfactant employed in the mechanical test.

## DISCUSSION

The use of in-plane swelling of guinea pig stratum corneum is an admittedly insensitive means of determining surfactant–skin interactions. Nevertheless, the procedure is simple, requires little time by an experimenter and no sophisticated equipment. That the values obtained for a given surfactant can differ with the source of the stratum corneum (animal to animal) and even with the site on an animal (piece to piece) is evident from the high standard deviations. Thus, in comparing surfactants, stratum corneum

from the same animal and from neighboring sites should be used. Fortunately, we have observed that although the magnitude may be variable from study to study, the relative order of magnitude remains the same and surfactants can easily be classified as producing little or no swelling, or producing readily measurable swelling. Scheuplein and Ross (4) soaked human stratum corneum in 5 per cent sodium laurate for up to 72 h and observed an expansion in area ranging from 50 to 80 per cent.

In discussing the mechanism by which anionic detergents swell stratum corneum while cationic and nonionic surfactants appear to have little effect, it seems reasonable to briefly consider the literature on globular proteins. As summarized by Tanford (9) amphiphilic substances (ionic or polar derivatives of hydrocarbons) can combine with proteins in at least 3 distinct modes of interaction. (1) Association with specific binding sites of native proteins (serum albumin and $\beta$-lactoglobulin behave in this manner); (2) Cooperative association between protein and a large number of detergent molecules without major conformational changes (serum albumin with alkyl sulfates and sulfonates having short hydrocarbon chains); (3) Cooperative association with gross denaturation of the protein. The 3 types of binding occur with detergent monomers rather than micelles. In fact, micelle formation can be considered to be in competition with protein binding. Nevertheless, to insure a maximum concentration of monomer, our studies were performed well above the CMC except where noted.

Since nonionic detergents have much lower CMC than anionic detergents, fewer monomers would be present in a solution of nonionic detergent so that binding type 3 would be less likely to occur with a nonionic detergent (10). It should be noted that little or no swelling was observed with nonionic detergents.

Tanford and coworkers (11) also compared the interaction of anionic lauryl sulfate and tetradecyltrimethylammonium ions with serum albumin and other globular proteins and concluded that both detergents yield type 3 interaction. However, with the cationic detergent type 3 binding occurred very close to its CMC while with lauryl sulfate the binding occurred far below its CMC. Less cationic detergent was bound than anionic detergent. The authors concluded that the difference between the 2 types of detergent was that the anionic detergent would cluster around the longer cationic sites in the protein (arginyl and lysyl side chains) which could accommodate more detergent molecules than the shorter anionic sites in the protein (glutamyl, aspartyl) around which cationic detergents would cluster. In a subsequent paper, however, Tanford and coworkers (12) studied the binding of the same cationic detergent as well as lauryl sulfate to apoproteins of human serum high density lipoprotein and observed cooperative interaction with the cationic detergent at a much lower equilibrium detergent concentration than observed previously with water-soluble proteins. They also concluded that binding the cationic and anionic detergent had resulted in a change in conformation which was unlike the denatured state observed with globular proteins. That little swelling was observed with the cationic surfactants we used might be due to the low CMC for Triton X-400, or it might reflect the inability of the stratum corneum to undergo cooperative binding with these particular surfactants. Preliminary work suggests that dodecyltrimethylammonium chloride may produce levels of swelling similar to lauryl sulfate. The results of the 2 studies just cited indicate the difficulty in choosing the appropriate soluble protein to simulate the insoluble stratum corneum. However, as the interests of academic scientists turn more toward membranous proteins, more choices for the cosmetic chemist/skin biologist should be available.

In reviewing the effects of chemicals on skin dimensions we should note the observation of Wildnauer that soaking stratum corneum in formic acid resulted in an increase in length of the tissue which he attributed to denaturation of the protein (13). Not all denaturing media induce this effect as indicated by our results shown in Table IV. Imokawa and coworkers (14, 15) used the change in optical rotation induced by soaking bovine serum albumin in several detergents as a means of estimating a detergent's ability to cause protein denaturation; they obtained positive correlation between a detergent's denaturing ability and its ability to produce "skin roughness" *in vivo*.

Scheuplein and Ross (4) observed that while concentrated urea solution had no effect on skin permeability to water, 1 per cent solutions of laurate and lauryl sulfate enhanced skin permeability. Interestingly, we observed no swelling in 8 *M* urea (Table IV), but notable swelling in the anionic surfactants. Not only was the swelling induced by laurate reversible ((4) and Table IV), but much of the barrier function was shown to be recoverable after resoaking the stratum corneum in water (4).

The observation of the reversibility of laurate-induced swelling and the observation that the birefringence of the stratum corneum, which is greatly diminished by soaking in 5 per cent laurate, is restored after resoaking in water led Scheuplein and Ross (4) to conclude that laurate had induced a reversible $\alpha \rightarrow \beta$ conversion of the stratum corneum protein with an uncoiling of the filaments, which was accompanied by a gross expansion of the tissue and high influx of water.

We conclude that denaturation does not cause stratum corneum swelling, but that swelling is due to a reversible conformation change resulting from cooperative binding of the appropriate detergent. The binding is neither type 2 nor type 3 but is probably most like that described above for the apoproteins of serum high density lipoproteins. Our observation on the effect of laurate, however, indicates irreversible effects as well. Prottey and Ferguson (16) have observed the extraction of proteins and amino acids by anionic detergents. Lauryl monoethoxysulfate can extract protein (16) but causes little swelling suggesting that protein extraction may be responsible for weakening but not for swelling.

Our results suggest that stratum corneum swelling could be of value for studying detergent-skin interactions and for predicting detergent penetration of skin and possibly subsequent skin irritancy.

## REFERENCES

(1) M. K. Polano, The interaction of detergents and the human skin, *J. Soc. Cosmet. Chem.*, 19, 3–20 (1968).
(2) B. R. Choman, Determination of the response of skin to chemical agents by an *in vitro* procedure, *J. Invest. Dermatol.*, 37, 263–71 (1961).
(3) P. W. A. Tovell, A. C. Weaver, J. Hope, and W. E. Sprott, The action of sodium lauryl sulfate on rat-skin-an ultrastructural study, *Brit. J. Dermatol.*, 90, 501–6 (1974).
(4) R. Scheuplein and L. Ross, Effects of surfactants and solvents on the permeability of epidermis, *J. Soc. Cosmet. Chem.*, 21, 853–73 (1970).
(5) E. K. Von Götte, Der einfluss von kolloidelectrolyten auf die Quellung isolierta epidermis, *Kolloid Z.*, 117, 42–7 (1950).
(6) E. J. Singer, L. J. Vinson, W. R. Koehler, M. D. Lehman, and T. Masurat, The nature of the epidermal barrier and some factors influencing skin permeability, *Toxicol. Appl. Pharmacol. Suppl. 2*, 7, 7–19 (1965).

(7) S. G. Elfbaum and M. A. Wolfram, Effect of dimethyl sulfoxide and other reagents upon mechanical properties of stratum corneum strips, *J. Soc. Cosmet. Chem.,* **21**, 129–40 (1970).
(8) T. V. Wachneldt, Sodium dodecyl sulfate in protein chemistry, *Bio. Systems,* **6**, 176–87 (1975).
(9) C. Tanford in *The hydrophobic effect: formation of micelles and biological membranes,* Wiley, New York, 1973, Ch. 15.
(10) S. Makino, J. A. Reynolds, and C. Tanford, The binding of deoxycholate and Triton X-100 to proteins, *J. Biol. Chem.,* **248**, 4926–32 (1973).
(11) Y. Nozaki, J. A. Reynolds, and C. Tanford, The interaction of a cationic detergent with bovine serum albumin and other proteins, *J. Biol. Chem.,* **249**, 4452–59 (1974).
(12) S. Makino, C. Tanford, and J. A. Reynolds, The interaction of polypeptide components of human high density serum lipoprotein with detergents, *J. Biol. Chem.,* **249**, 7379–82 (1974).
(13) G. L. Wilkes, I. A. Brown, and R. H. Wildnauer, The biomechanical properties of skin, *CRC Critical Rev. in Bioengineering,* 453–95 (1973).
(14) G. Imokawa, K. Sumura, and M. Katsumi, Study on skin roughness caused by surfactants: correlation between protein denaturation and skin roughness, *J. Amer. Oil Chem. Soc.,* **52**, 484–9 (1975).
(15) G. Imokawa, K. Sumura, and M. Katsumi, Study on skin roughness caused by surfactants: a new method *in vivo* for evaluation of skin roughness, *J. Amer. Oil Chem. Soc.,* **52**, 479–83 (1975).
(16) C. Prottey and T. Ferguson, Factors which determine the skin irritation potential of soaps and detergents, *J. Soc. Cosmet. Chem.,* **26**, 29–46 (1975).
(17) P. Mukerjee and K. Mysels, Critical micelle concentration of aqueous surfactant systems, NSRDS-NBS 36 (1971).