J. Soc. Cosmetic Chemists, **16**, 783–794 (1965)

Identification of Surface Active Agents As Trimethyl Silyl Ether Derivatives by Gas Chromatography

ROBERT SUFFIS, M.A., THOMAS J. SULLIVAN, B.S., and WILLIAM S. HENDERSON, B.S.*

Presented May 4, 1965, New York City

Synopsis—A method is presented for the analysis of some non-ionic surface active agents by gas chromatography. The components of these agents are converted to their volatile trimethyl silyl ether derivatives prior to analysis by reaction with hexamethyldisilazane and trimethylchlorosilane. The volatile derivatives of the surface active agents may then be easily separated by gas chromatography. This procedure has been found to be applicable to a variety of glycol esters and sorbitan esters which are frequently utilized in cosmetic and toiletries formulations. In addition, the method could be utilized to provide information concerning the chemical properties of a surface-active agent. Rapid analysis for mono-ester and di-ester concentrations, free glycol, and fatty acid composition is possible through use of this technique.

Introduction

The analysis of partial esters of polyhydric alcohols and other non-ionic surface active agents has been performed by chromatographic techniques. These methods have utilized silica gel columns and various solvent systems to effect these separations (1–3). In addition, there is considerable literature on the analysis of glycerides by paper (4, 5) and thin-layer chromatography (6–8). Research on these separations has also been performed utilizing countercurrent distribution (9) and liquid-liquid extraction (10). Most of the work in this field has been per-

* The Mennen Company, Morristown, N. J.

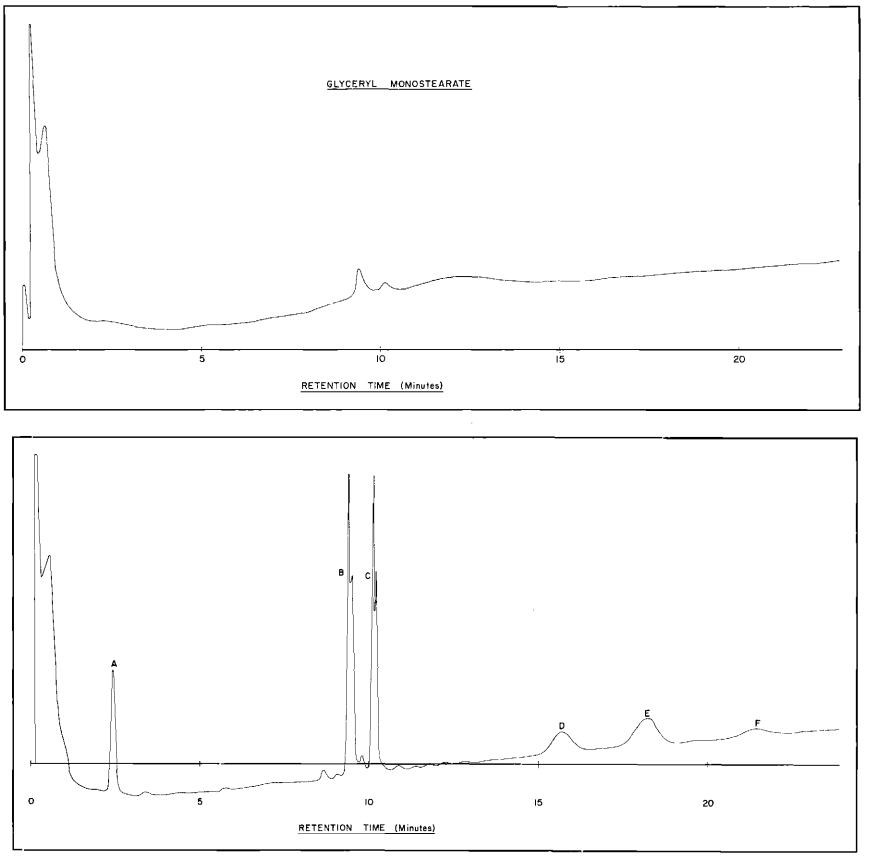

Figure 1. Top: Glyceryl monostearate in pyridine; Bottom: Glyceryl monostearate after trimethylsilylation

formed by lipid chemists. Therefore, most of the data cover fatty glycerides only. However, the same approaches should be possible for any glycol ester.

The above references describe techniques for the separation of the mono-ester, di-ester, and tri-ester components of the glycerides. These methods, applied to the identification of surface active agents, are lengthy, tedious, and usually not sufficiently specific for unequivocal identification.

There has been some research into the use of gas chromatography for the analysis of glycol esters. Triglycerides have been analyzed directly by high-temperature gas chromatography (11). However, the mono and diglycerides cannot be analyzed without conversion to a non-polar

derivative. The high boiling point and high polarity caused by the presence of one or more hydroxyl groups make it impossible to get good results by gas chromatography on the parent substance. Monoglycerides have been analyzed after conversion to allyl esters by a dehydration reaction (12). This method is applicable to glyceryl mono-esters only. Other work has been performed after converting the free hydroxyl groups

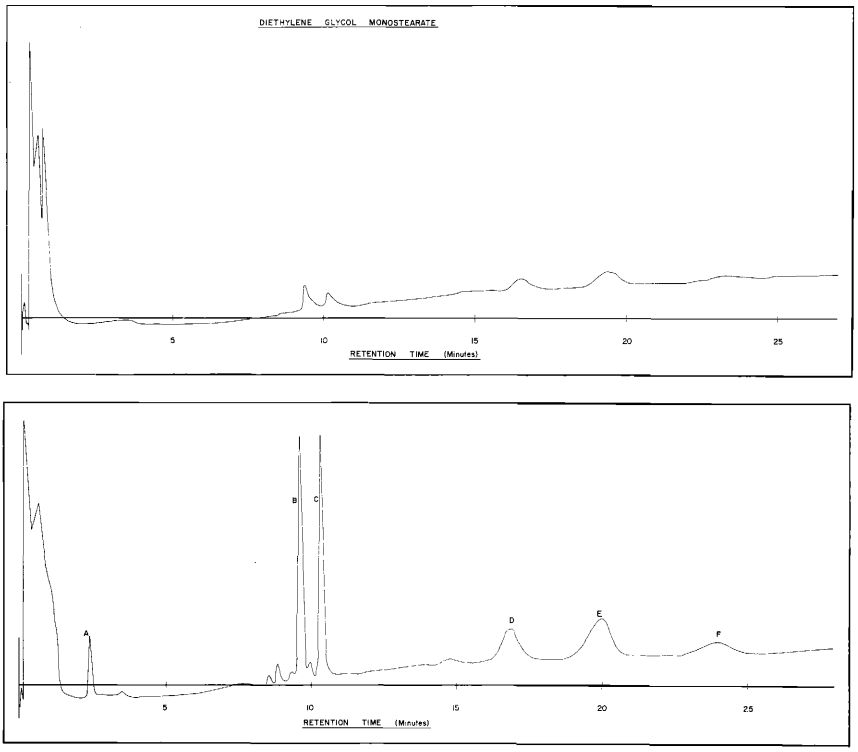

Figure 2. Top: Diethylene glycol monostearate in pyridine; Bottom: Diethylene glycol monostearate after trimethylsilylation

to acetates by reaction with acetyl chloride (13). These techniques have given some interesting results, but each one has some serious drawbacks. The conversion to allyl derivatives is very lengthy and applicable only to components with two adjacent hydroxyl groups. The acetate derivative preparation is also a lengthy procedure and gives rise to high boiling products.

The research described in this paper has made use of the reaction of hydroxyl groups contained in surface active agents with hexamethyl-

disilazane and trimethylchlorosilane. The products are volatile trimethyl silyl ether derivatives which are suitable for gas chromatography. The use of this reaction has received considerable attention in the sugar and carbohydrate field (14, 15). Compounds as high boiling as tetrasaccharides have been analyzed successfully. The reaction is simple and quantitative without any undesirable side reactions.

These derivatives are lower boiling than the corresponding acetates and give sharp, well defined peaks on the gas chromatograph. The use

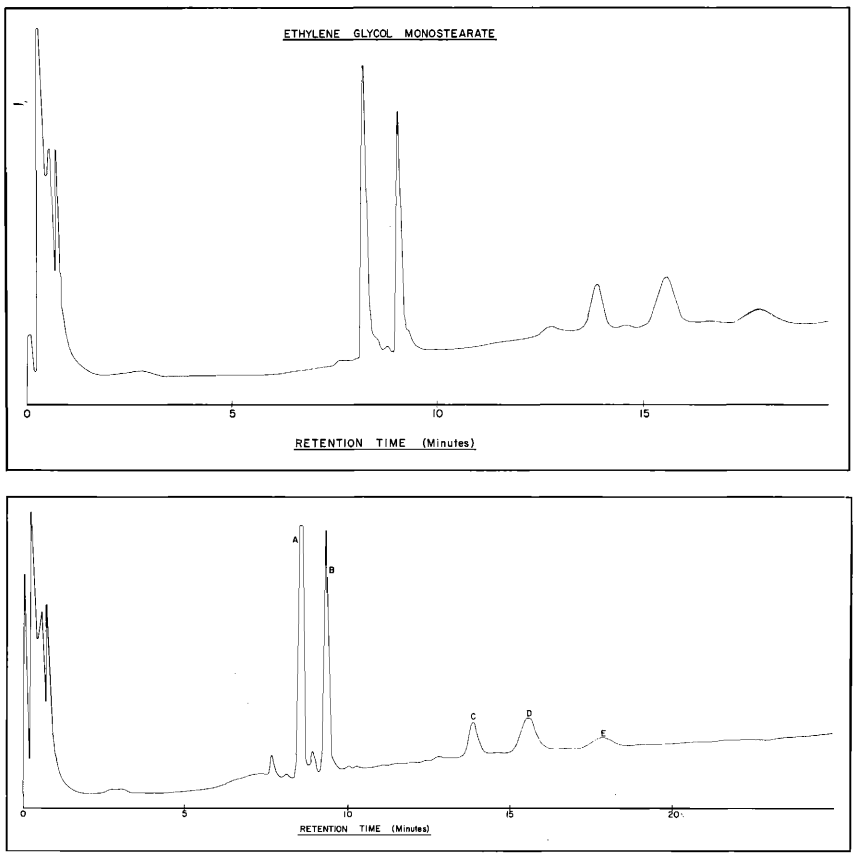

Figure 3. Top: Ethylene glycol monostearate in pyridine; Bottom: Ethylene glycol monostearate after trimethylsilylation

of this technique enables one to get a definitive chromatogram for each of the surface active agents investigated. A comparison of the chromatogram obtained from the initial pyridine solution with that of the reacted

product provides additional criteria for identification. This may be done easily by first analyzing a sample of the surface active agent in pyridine. Then the two reagents can be added and the chromatogram of the trimethylsilylated derivatives run next.

Experimental

Preparation of Derivatives

About 50–100 mg. of surface active agent is dissolved in 1 ml. of anhydrous pyridine (kept over KOH pellets) in a small plastic stoppered vial, and 0.2 ml. of hexamethyldisilazane and 0.1 ml. of trimethylchlorosilane are then added. The mixture is shaken vigorously for 30 seconds and then allowed to stand for 5 minutes. The solutions become cloudy, and a precipitate of ammonium chloride is formed. It is not necessary to remove this precipitate. The supernatant liquid may be run directly by gas chromatography (14).

Gas Chromatography

An F&M Model 810 Gas Chromatograph with thermal conductivity detector was used. All of the chromatograms were run using identical conditions. A 3 ft. by 0.125 in. o.d. column packed with 5% SE-52 silicone gum rubber on Anakrom A was employed. The helium flow rate was 20 ml./minute, and the column temperature was programmed from 100 to 300°C at a rate of 20°C/minute. One-microliter samples were injected into the gas chromatograph, using a Hamilton 10 microliter syringe.

Results and Discussion

Figure 1 shows a comprison of the chromatograms obtained from glyceryl monostearate before and after trimethylsilylation. The top chromatogram was obtained from a solution of glyceryl monostearate in pyridine. It is apparent that very little information can be obtained from this. The high boiling point and polar character of these components show very pronounced tailing for those peaks that have passed through the column. The bottom chromatogram shows the results obtained for the glyceryl monostearate derivatives after trimethylsilylation. The significant improvement is apparent.

All of the major peaks have been identified. Peak A is glycerin, and Peaks B and C are glyceryl mono-esters of palmitic and stearic acids, respectively. The small peak in front of peak B is due to glyceryl

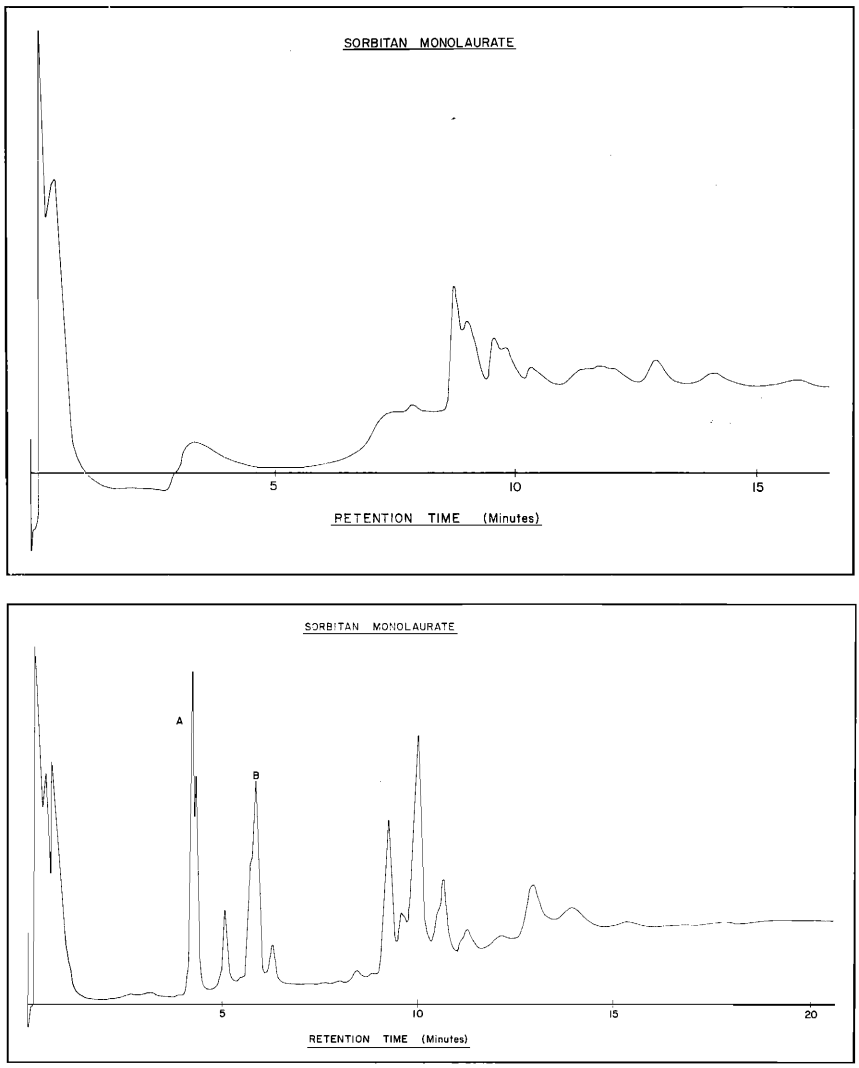

Figure 4. Top: Sorbitan monolaurate in pyridine; Bottom: Sorbitan monolaurate after trimethylsilylation

monomyristate. The composition of the monoesters is indicative of the fatty acid composition of the glyceryl ester. Peaks D, E, and F are diester peaks. Peak D is the dipalmitate; peak F is the distearate. The peak between these is due to the mixed ester of palmitic and stearic acids. These three peaks have been found to be in the expected ratio for the random combination of stearic and palmitic acids with glycerin. This

chromatogram together with the chromatogram of the glyceryl monostearate in pyridine provide for an absolute identification of this material.

Glyceryl esters of other fatty acids are also readily identifiable by this technique. It is simple to distinguish glyceryl monostearate prepared

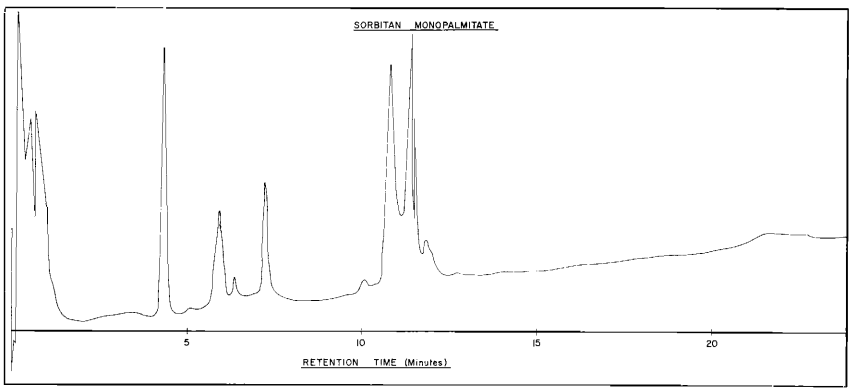

Figure 5. Sorbitan monopalmitate (Arlacel 40) after trimethylsilylation

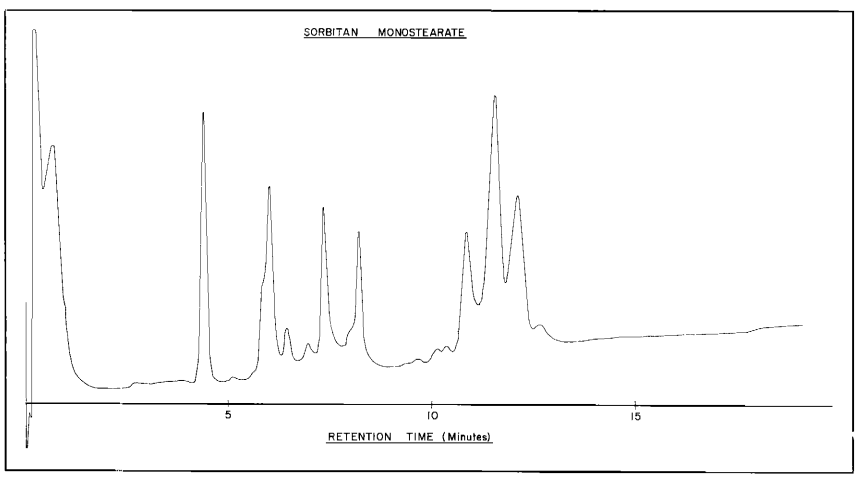

Figure 6. Sorbitan monostearate (Arlacel 60) after trimethylsilylation

from pressed stearic acid and that prepared from hydrogenated tallow fatty acids. Under the conditions of analysis utilized in this study there is no separation of the α- and β-monoglycerides. However, according to a recent publication (16) it is possible to separate these compounds using an ethylene glycol succinate column. This column has the disadvantage

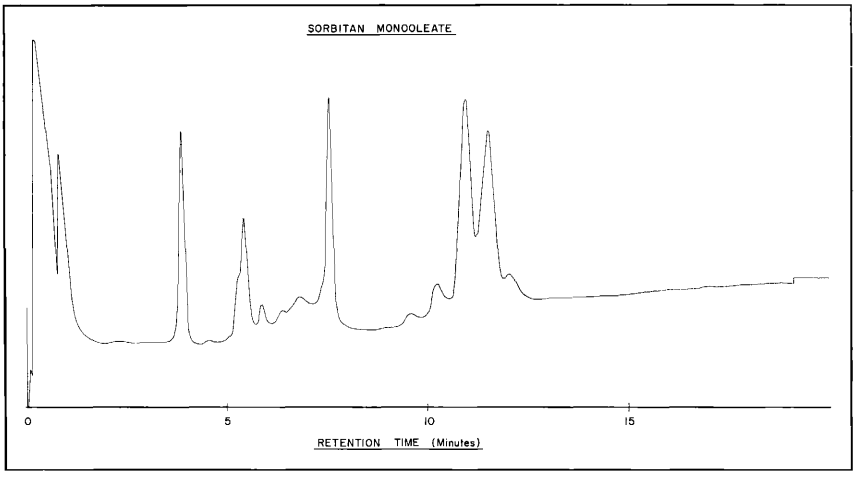

Figure 7. Sorbitan mono-oleate (Arlacel 80) after trimethylsilylation

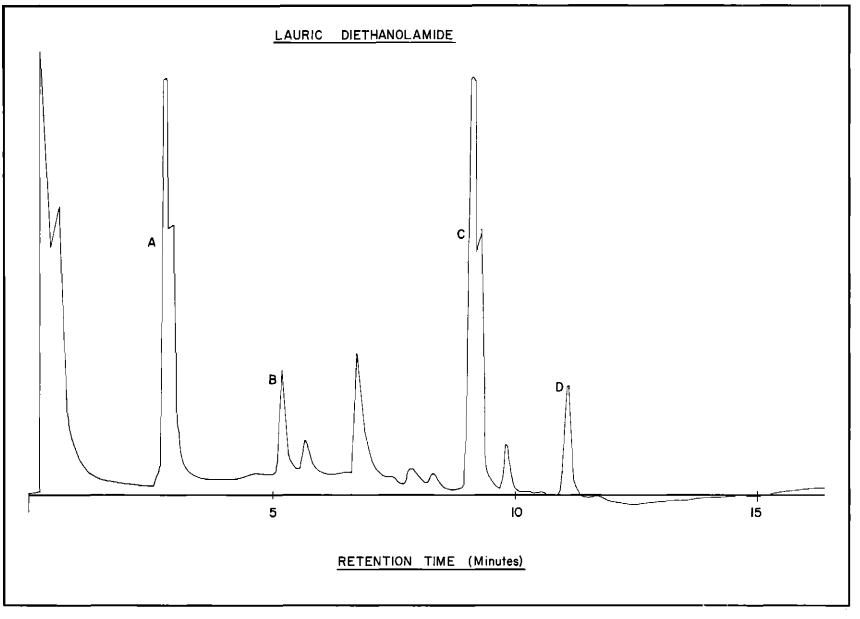

Figure 8. Lauric acid diethanolamide after trimethylsilylation

of comparatively low temperature stability, so it cannot be used for analysis of the di-ester components.

Figure 2 shows a comparison of the chromatograms of diethylene glycol monostearate before and after trimethylsilylation. One immediately notices the fact that peaks D, E, and F have identical retention

times in both chromatograms. These peaks are assigned to the di-esters present in diethylene glycol monostearate. These compounds have no available hydroxyl groups and, therefore, do not react with hexamethyldisilazane. The fact that these components are identical in the two chromatograms confirms the fact that this is a mixture of the di-esters derived from diethylene glycol. The mono-ester components, peaks B and C, which still contain a hydroxyl group do show a considerable change on trimethylsilylation. Peak A is due to free diethylene glycol.

Figure 3 shows a comparison of the chromatograms for ethylene glycol monostearate before and after reaction with hexamethyldisilazane. In this case, the di-ester peaks C, D, and E are identical in both chromatograms. Peaks A and B, due to the mono-ester, are also almost the same in both cases. This is due to the relatively non-polar characteristic of the mono-ester of ethylene glycol monostearate. However, there are changes that have occurred in the mono-ester peaks, before and after trimethylsilylation. The retention times of the two unreacted components are slightly longer than those of their counterparts in the bottom chromatogram. In addition, the reacted mono-esters show a considerably greater response for the same sample size. Even though the peaks have not undergone the considerable changes shown in the other examples, it is still apparent that some reaction has taken place. There is no free ethylene glycol peak, since the derivative of this component is so low boiling it is not resolved from the pyridine used as a solvent.

The three glycol stearates shown thus far can be easily distinguished by their chromatograms. Not only is there a difference between their response before and after trimethylsilylation, but in addition there are differences in the retention times of the various components which provide for absolute identification.

It can be readily seen that through the use of calibration standards it should be possible to analyze these glycol stearates quantitatively for free glycol, mono-esters, and di-ester concentration. Presently used methods for glycol and mono-ester are rather lengthy wet chemical procedures. There is no direct quantitative method for di-ester content commonly in use. The di-ester content could be an important factor in the performance of a surface active agent, such as glyceryl monostearate

Other glycol esters that have been studied by this procedure include diethylene glycol mono-oleate, propylene glycol monostearate, diethylene glycol monolaurate, diglycerol monostearate, triglycerol monostearate, and decaglycerol monostearate. In each case the surface

TABLE I

Relative Retention Times of Glycolesters and Sorbitan Esters (Glyceryl Monopalmitate = 1.000)

	Glycerol Mono-Stearate	Ethylene Glycol Monostearate	Diethylene Glycol Monostearate	Diethylene Glycol Monooleate	Propylene Glycol Monostearate	Diethylene Glycol Monolaurate	Decaglycerol Mono-Stearate	Diglycerol Mono-Stearate	Triglycerol Mono-Stearate
Free Glycol									
A	0.257		0.259	0.259		0.217	0.247	0.264	0.247
B							0.452	0.622	0.446
C							0.531		0.608
D							0.613		
Monoester									
A	1.000	0.898	1.026	0.866	0.804	0.833	0.774	0.860	0.769
B	1.077	0.983	1.104	1.098	0.901	0.920	0.849	1.013	0.854
C					0.983	1.004	0.987	1.095	1.011
D						1.075	1.036	1.188	1.091
E								1.253	1.188
F									1.258
Diester									
A	1.659	1.466	1.794	1.759	1.339	1.222	1.183	1.348	1.344
B	1.928	1.650	2.123	2.053	1.458	1.295	1.328	1.454	1.462
C	2.269	1.903	2.540	2.487	1.643	1.387	1.587	1.688	1.753
D					1.858	1.518		1.962	1.989
E						1.716		2.344	2.389
F								3.022	3.064
G									3.914

active agent could be readily identified by its chromatogram, which was distinguishable from any other gylcol ester.

Several sorbitan esters have been investigated by this technique. Figure 4 shows a comparison of the chromatograms for sorbitan monolaurate (Arlacel 20) before and after trimethylsilylation. The Arlacel 20 is broken down into a large number of components. Peaks A and B represent the main components of free sorbitan. Peak A is probably the hexide or five-membered ring dehydration product of sorbitol. Peak B has been assigned as the hexitan or six-membered ring dehydration product. This pattern is repeated with some minor modifications in each of the sorbitan ester combinations.

In the example shown, the fatty acid component is lauric acid. No attempts have been made thus far to make any definite assignments to these peaks. For the purpose of this investigation it is significant to note the differences among the sorbitan esters and use these data as a method of identification. The differences between Arlacels 20, 40, 60, and 80 are apparent after comparison of Figs. 5, 6, and 7 with Fig. 4.

It is also apparent that this gas chromatographic method could be an important tool in the elucidation of the composition of these surface-active agents. Table I gives a listing of the relative retention times of the major peaks of each of the glycol esters and sorbitan esters run by this technique. In each case there is no problem in identification of the surface active agent from its definitive chromatogram.

The trimethylsilylation reaction has also been applied to several other cosmetic raw materials. Figure 8 shows a chromatogram of lauric acid diethanolamide after trimethylsilylation. Peak A in this chromatogram is due to free diethanolamine, peak B to free lauric acid, and peak C (the main peak) to the amide. Peak D has been identified as the amine ester. The minor peaks have not been identified. Further investigation of this raw material may provide for identification of the amide ester that is usually present in this raw material. Possibly other more selective columns might perform an improved resolution job. The use of this technique for analysis of ethanolamines is another important application. The diethanolamine impurity gives an excellent peak (Fig. 8). Equally sharp, well resolved peaks are given by monoethanolamine and triethanolamine.

Summary

The technique of trimethylsilylation followed by gas chromatography appears to be applicable to the identification of some non-ionic surface-

active agents. It has been found to be particularly useful for glycol esters and sorbitan esters. Some of the possibilities of extension of this work to quantitative analysis of the components of these surface-active agents has been mentioned. In addition, fatty amides and ethanolamines have been indicated as areas of research that would merit further interest.

(Received July 16, 1965)

References

(1) Hofmann, A. F., *J. Lipid Res.*, **3,** 391 (1962).
(2) Hamilton, J. G. and Holman, T. T., *J. Am. Chem. Soc.*, **76,** 4107 (1954).
(3) Huebner, V. R., *J. Am. Chem. Soc.*, **35,** 325 (1958).
(4) Holasek, A., and Fried, J., *Mikrochim Acta*, **1957,** 469.
(5) Dieckert, J. W., and Reiser, R., *J. Am. Oil Chem. Soc.*, **33,** 123 (1956).
(6) Privett, O. S., and Blank, M. L., *Ibid.*, **39,** 520 (1962).
(7) Rybecka, S. M., *Chem. Ind.* (London) **1962,** 308.
(8) Vioque, E., and Holman, R. T., *J. Am. Oil Chem. Soc.*, **39,** 63 (1962).
(9) Perry, E. S., and Brokow, G. Y., *Ibid.*, **32,** 652 (1955).
(10) Monick, J. A., and Treybal, R. E., *Ibid.*, **33,** 193 (1956).
(11) Fryer, F. H., Ormand, W. L., and Crump, G. B., *Ibid.*, **37,** 589 (1960).
(12) McInnes, A. G., Tattrie, N. H., and Kater, M., *Ibid.*, **37,** 7 (1960).
(13) Huebner, V. R., *Ibid.*, **36,** 262 (1959).
(14) Sweeley, C. C., Bentley, R., Makita, M., and Wells, W. W., *J. Am. Chem. Soc.*, **85,** 2497 (1963).
(15) Bentley, R., Sweeley, C. C., Makita, M., and Wells, W. W., *Biochem. Biophys. Research Commun.*, **11,** 14 (1963).
(16) Wood, R. D., Raju, P. K., and Reiser, R., *J. Am. Chem. Soc.*, **42,** 161 (1965).

ERRATUM

Due to language difficulties an unavoidable error occurred in the synopsis of "Approaches to a Prophylaxis of Skin Aging," by M. and H. Ippen, *J. Soc. Cosmetic Chemists*, **16,** 305-8 (1965). The corrected synopsis should read as follows:

Synopsis—It is shown that smoking has a deleterious effect on skin condition and that this effect can be differentiated from that of damage by sunlight. Smoker's skin is identified as skin which suffers from loss of "turgor" and shows signs of flabbiness; in addition, the color of the smoker's skin is pale, with a grayish hue. Dermatological examination of 224 women up to now show moderate correlation between their smoking habits and the appearance of their skin, as defined above. By contrast, smoking seems to have only a very minor effect on the skin of male smokers.

For the convenience of the readers, the corrected synopsis is repeated on page xxxvii for use in card indexes.