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Note

Determination of carbon chain distribution in alkyl sulfates by *in situ* hydrolysis–gas chromatography

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Sodium dodecyl sulfate (SDS) is routinely used in gel electrophoresis and as a reagent to solubilize proteins. It is now recognized that many commercial samples of SDS are contaminated with homologous alkyl sulfates, mainly tetradecyl sulfate (STS)[1–3]. Dohnal and Garvin[2] reported that STS can alter the affinity of proteins to certain stains and that STS shifts monomer–dimer equilibria of proteins to the monomer form. Thus there is a need for a convenient method for the analysis of SDS. In this paper, we will report a facile method by which the carbon chain distribution in SDS samples may be rapidly determined. SDS samples in water are injected directly into the gas chromatograph with no sample pretreatment and undergo quantitative *in situ* conversion to the corresponding alcohols which are separated and detected.

The determination of the carbon chain distribution in commercial samples of SDS has been accomplished by several methods. All of these methods require sample pretreatment. For example, acid hydrolysis for 1–3 h yields a mixture of homologous fatty alcohols which are extracted into a solvent, *e.g.*, diethyl ether or hexane[1,4]. The solvent final volume is then adjusted prior to analysis by gas chromatography (GC). Pyrolysis–GC of SDS furnishes a mixture of products, primarily 1-olefins and fatty alcohols from which the carbon chain distribution may be derived[5]. Pyrolysis over KOH results in a mixture of 1-olefins with lesser yields of internal olefins and dialkyl ethers which are swept into the gas chromatograph and detected[6]. From pyrolysis–GC over P_2O_5 or H_3PO_4, a mixture of olefins is obtained[7].

EXPERIMENTAL

Materials

Decanol, dodecanol, tetradecanol and 1-dodecene were obtained from Aldrich and were 99% pure by GC. SDS and STS were obtained from Eastman-Kodak. Sodium dodecyl sulfate was obtained from Eastman-Kodak, Fisher Scientific, Pierce, Polysciences, and Sigma. Lithium dodecyl sulfate was obtained from Polysciences and Sigma. Solutions of fatty alcohols were prepared in reagent-grade methanol. Solutions of alkyl sulfates were prepared in distilled, deionized water.

Methods

GC analyses were made with a Varian Aerograph Model 3700 under the fol-

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lowing conditions: injector temperature, 270°C; flame ionization detector, 230°C; attenuation, $128 \cdot 10^{-10}$ A/mV; helium flow-rate, 20 ml/min; column temperature, 200°C. The following 6 ft. × 1/8 in. stainless-steel columns were used: 10% SP-2100 on 80–100 mesh Supelcoport and 5% Carbowax 20M on Anakrom SD. PTFE-coated Microsep septa were utilized. Sample injection volumes of 1, 2 or 3 μl were delivered with a Precision 5-μl syringe. Quantitation of peak areas was accomplished by cutting out the peaks and determination of peak mass with a Mettler H31 AR balance. The elution patterns for aqueous solutions of SDS and methanolic solutions of decanol,

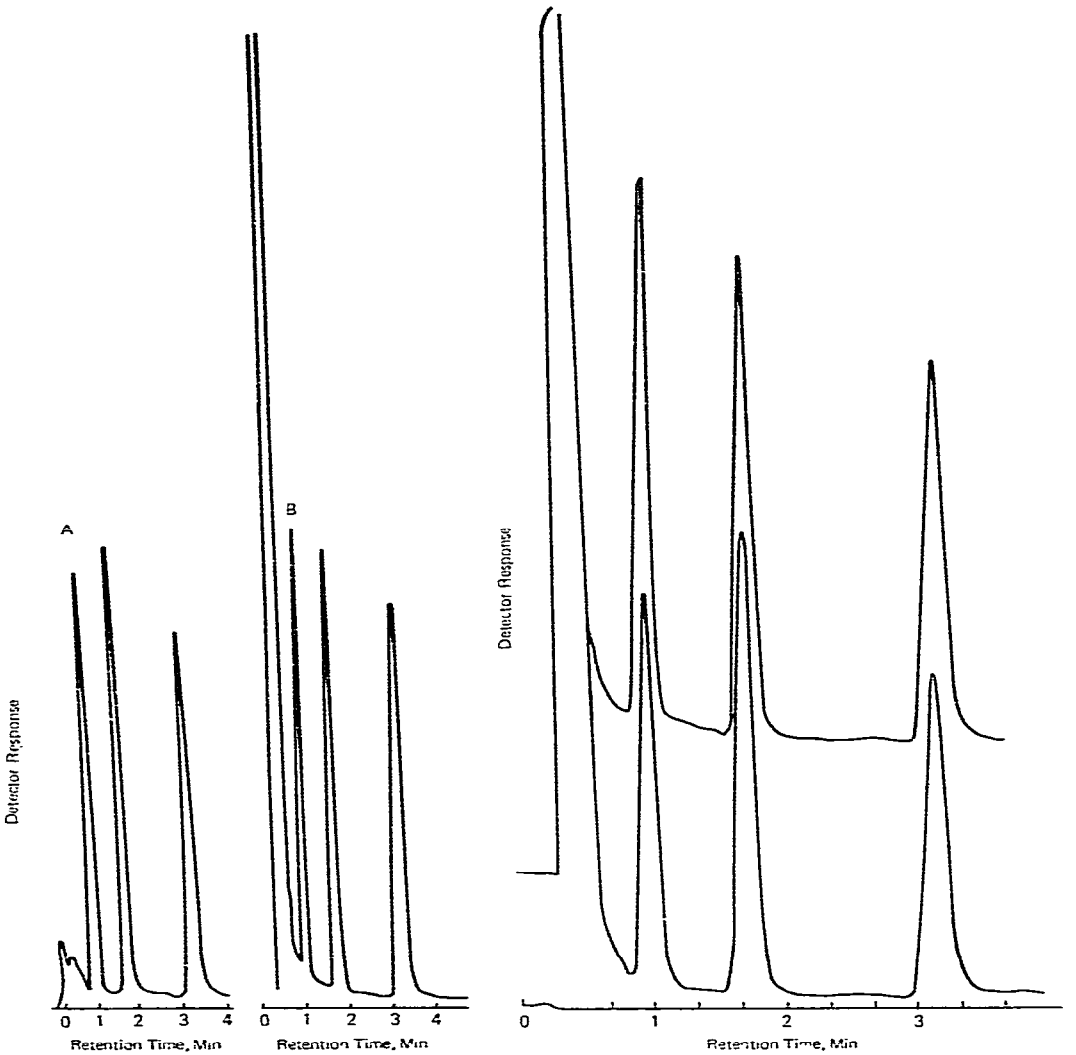


Fig. 1. A, Equimolar 0.026 *M* solution of C_{10}, C_{12}, C_{14} alkyl sulfates in water. B, Equimolar 0.026 *M* solution of C_{10}, C_{12}, C_{14} alcohols in methanol, SP-2100 column. In both series, the order of elution is C_{10}, C_{12}, C_{14}.

Fig. 2. Top, equimolar 0.026 *M* solution of C_{10}, C_{12}, C_{14} alcohols in methanol. Bottom, equimolar 0.026 *M* solution of C_{10}, C_{12}, C_{14} alcohols plus SDS in water, SP-2100 column.

dodecanol and tetradecanol were very reproducible throughout the course of this work. The SP-2100 column was baked at 300°C briefly at the beginning and end of each work day. A glass-lined injector packed with glass wool was used.

RESULTS AND DISCUSSION

It has been found that on an SP-2100 column, the chromatogram of an equimolar mixture of decanol, dodecanol, and tetradecanol in methanol yields three peaks with the identical retention times that are observed for an equimolar mixture of C_{10}, C_{12}, and C_{14} alkyl sulfates in water (see Fig. 1). In Fig. 2, the fatty alcohol mixture has been diluted with an aqueous solution of C_{12} sulfate. The C_{12} peak, at 1.7

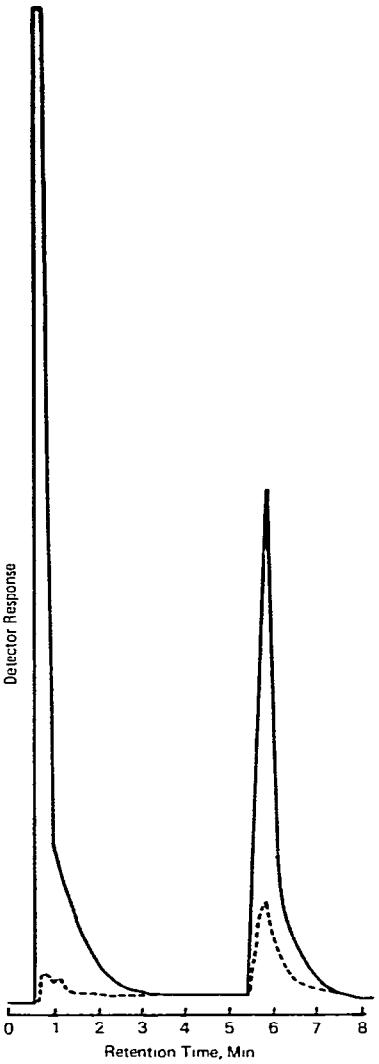


Fig. 3. ———, 0.026 *M* Dodecanol in methanol; ·····, 0.003 *M* SDS in water, Carbowax 20M column.

min, is enhanced and exhibits no shoulders. The basis for chromatographic separation on silicone polymer columns is related to difference in boiling point[8]. Since dodecanol boils at 260°C and SDS melts at 204–207°C, the alkyl sulfate is presumably converted to the corresponding alcohol in the injector. Dodecanol in methanol and SDS in water also yield the same retention time (5.8 min) on a Carbowax 20M column which separates compounds by differences in polarity[8] (see Fig. 3). SDS is not converted to the 1-olefin since 1-dodecene has a retention time of 0.6 min on SP-2100.

Next, the quantitative response (per mole) was determined for dodecanol and SDS and analogously for tetradecanol and STS. Calibration curves for the C_{12} and C_{14} alcohols were prepared by injection of 1-, 2- and 3-μl volumes of an equimolar 0.026 *M* solution in methanol. Peak area determination by cutting and weighing gave superior results to either triangulation or peak height. Significantly, 3 μl of an aqueous equimolar 0.026 *M* solution containing Polysciences SDS and Eastman-Kodak STS yielded the same detector response as the corresponding alcohol (see Fig. 4). This shows that the chromatograms obtained for the alkyl sulfates are a result of a total *in situ* conversion of the sample in the GC injector.

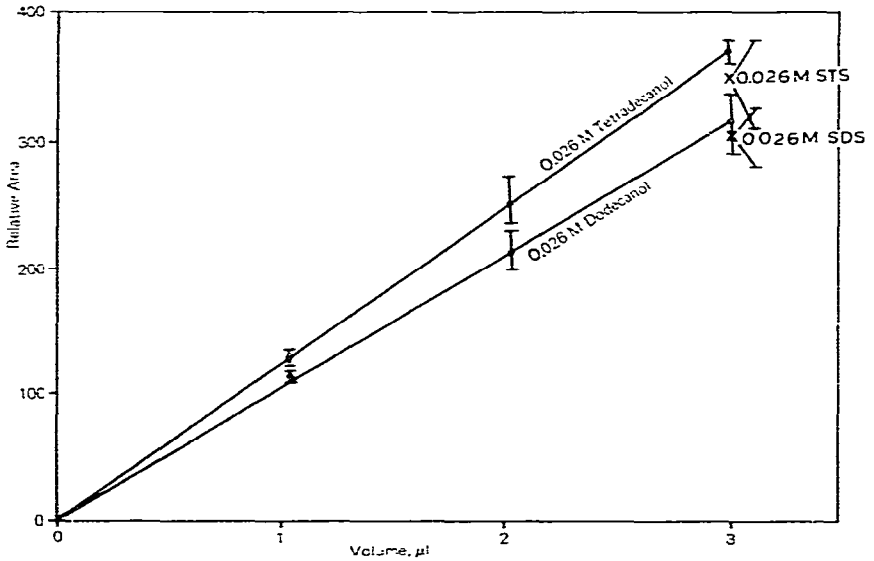


Fig. 4. Quantitative response for tetradecanol and STS (upper line) and dodecanol and SDS (lower line). All data points are the mean of 5 replicates; error bars are ± standard deviation. SP-2100 column.

It has been reported that commercially available samples of SDS are frequently found to contain 25–31 % of STS[1–3]. In Table I are shown the results of carbon chain distribution analysis of commercially available samples of SDS as determined by the direct injection technique. It is apparent that there is a wide range of purity among these samples. The major impurity is STS. It is noted that Pierce SDS yields different results by direct injection *vs.* hydrolysis and extraction. However, the probable cause of this apparent discrepancy is that these SDS samples represent different lots of material. The observed chain distribution for Sigma SDS agrees quite well with the label values.

TABLE I

CARBON CHAIN DISTRIBUTION IN COMMERCIAL ALKYL SULFATES

SDS = Sodium dodecyl sulfate; LDS = lithium dodecyl sulfate; STS = sodium tetradecyl sulfate.

Source	*Alkyl sulfate*	*Percent total area**			
		C_{10} (%)	C_{12} (%)	C_{14} (%)	C_{16} (%)***
Eastman-Kodak, Lot No. A0A	SDS	0.45	99	0.33	–
Fisher Scientific, Lot No. 702789	SDS	2.1	65	26	7.9
Pierce, sequanal grade, Lot No. 021981-9	SDS	0.43	98	1.1	–
Pierce**	SDS	–	99	0.56	–
Polysciences, Lot No. 2-1178	SDS	0.38	98	1.5	–
Polysciences, Lot 04098	LDS	0.85	99	–	–
Sigma, Lot No. F0F-0302	SDS	0.68	67	27	5.6
Sigma, Lot No. F0F-0302, label values	SDS	–	65	27	6
Sigma, Lot No. 11F-0056	LDS	0.80	99	–	–
Eastman-Kodak, Lot No. B9A	STS	–	5.0	95	–

* Percentages computed by cut and weigh procedure; values not corrected for molar response factor. Precision for duplicate injections ±1%. Sample concentration, 2.0% in water; sample volume, 3.0 μl.

** Determined by injection after acid hydrolysis and extraction[4].

*** The data for % C_{16} in SDS samples is based on the presence of peaks with a retention time of 6.0 min which is also observed for hexadecanol in methanol. Hexadecyl sulfate is not commercially available.

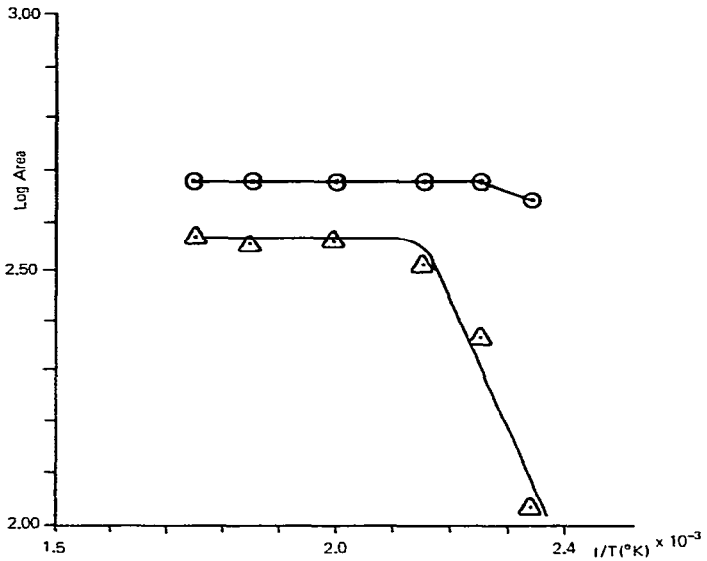
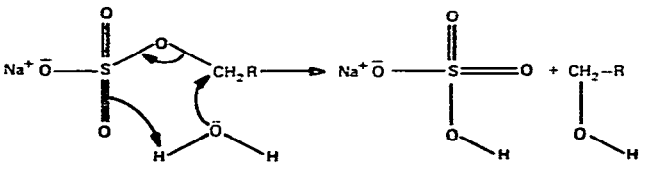


Fig. 5. Effect of injector temperature (T) on peak area for 0.035 M dodecanol in methanol (○) and 0.026 M SDS in water (△), SP-1200 column.

The effect of injector temperature on peak area was compared for 0.035 *M* dodecanol in methanol and 0.026 *M* SDS in water. The effect of temperature from 150–300°C was negligible on dodecanol. In contrast, over the same temperature range, the peak area for SDS markedly diminished as the injection temperature was lowered. The data were transformed and plotted as log area *vs.* $1/T$ (see Fig. 5). From this plot, an activation energy of 11 kcal/mole was calculated.

The gas-phase hydrolysis of the alkyl sulfate must occur within the confines of the injector and therefore must be extremely fast. A concerted mechanism involving a cyclic intermediate is proposed:



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