Chemical and Physical Properties of Isomeric Glyceryl Monoethers[1]

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ABSTRACT

Long-chain saturated and mono- and diunsaturated 1- and 2-glyceryl monoethers were synthesized by reacting 1,2-isopropylidene and 1,3-benzylidene glycerol potassium salts with alkyl halides in the preparation of the saturated monoethers, and with alkenyl-*p*-toluenesulfonates in the preparation of the unsaturated monoethers, followed by hydrolysis of the blocking groups with boric acid. The progress of the reaction was monitored by gas-liquid chromatography (GLC) of the reaction mixture. The 2-glyceryl ethers, with two exceptions, had not been prepared previously. Normal propyl and 3-pentyl octadecyl ethers also were synthesized to aid in the interpretation of infrared (IR) and nuclear magnetic resonance (NMR) spectra. All the ethers prepared were purified by preparative thin-layer chromatography (TLC) and crystallization. Their purity was found to be greater than 95%, as determined by TLC and GLC, supported by NMR and IR spectra. The isomeric 1- and 2-glyceryl ethers were separated on Silica Gel G adsorbent layers impregnated with either sodium arsenite or boric acid and their TLC behavior interpreted, based on the polarity of the complexes formed. Melting point determinations indicated more than one polymorphic form. Comparison of IR and NMR spectra of the saturated and unsaturated isomeric glyceryl ethers, and various derivatives, demonstrated the applicability of these spectroscopy methods for characterization and structural determination, in addition to distinguishing between the two isomeric forms.

INTRODUCTION

GLYCERYL ETHERS CAN EXIST in two isomeric forms: The hydrocarbon chain can be linked to the one-position of the glycerol molecule (1-isomer) or the two-position (2-isomer). The former is unsymmetrical and can exist as either of two enantiomeric forms, but the latter is symmetrical and can exist in only one form. Heilbron and co-workers, in a series of investigations (1-4), established that the naturally occurring glyceryl ethers found in the oils of elasmobranch fish are the 1-isomer. Baer et al. (5-7) demonstrated by comparison with synthetic enantiomers of the 1-glyceryl ethers that the naturally occurring ethers found in elasmobranch fish oils possessed the D configuration.

Since these early investigations, glyceryl ethers, which were once thought to be exclusively associated with the oils of various marine animals, have been found in many mammalian tissues and fluids, vegetable oils, and domestic hen eggs (8-21) as diesters and phosphatides. Owing to the low concentrations in which the glyceryl ether usually occurs in mammalian tissues and fluids, many investigators assumed without adequate proof that only the 1-isomer was present. The inability to distinguish between the two isomers with microgram quantities reflects the inadequacy of the present analytical methods. Until recently (22), a method did not exist for the simultaneous determination of the 1- and 2-glyceryl ethers in a mixture. The recorded physical and chemical properties of the 1-glyceryl ethers are scant, and the purity of the material used to make the determinations in some cases is questionable. Baumann and Mangold (33) have prepared a number of 1-isomers in a high state of purity and have determined their melting points and critical solution temperatures. The physical and chemical properties of the 2-glyceryl ethers, like the 1-isomer, are even more inadequately known, since only two of these isomers have been prepared and purified.

This lack of basic knowledge of the isomeric glyceryl ethers led us to make a detailed comparison of the physical and chemical properties of several purified synthetic saturated and mono- and diunsaturated 1- and 2-glyceryl ethers and some of their derivatives. The results obtained by comparison of infrared (IR) and nuclear magnetic resonance (NMR) spec-

[1]Presented at the AOCS Meeting, Philadelphia, October 1966.

[2]An operating unit of the Oak Ridge Associated Universities, under contract with the US Atomic Energy Commission.

tra, melting points, and gas-liquid chromatography (GLC) retention times for both isomers of the various glyceryl ethers are reported here.

ANALYTICAL METHODS

Analytical Gas Chromatography

The gas-chromatographic system and operating parameters used in this investigation have previously been described (22). Column specifications and operating conditions used for the analysis of the isomeric glyceryl ethers, starting materials, and synthetic intermediates are shown in Table I. Columns were packed, tested, and conditioned thermally and with trifluoroacetic anhydride before use (22). The information obtained from the GLC analyses of the compounds shown in Table I, other than the isomeric glyceryl ethers, which have been shown to give quantitative results (22), was used in a qualitative or semiquantitative manner only. Quantitative results were calculated from peak area measurements determined by triangulation.

Preparative Gas Chromatography

An Aerograph Model 90-P3 gas chromatograph (Wilkens Instruments and Research, Inc., Walnut Creek, Calif.), equipped with a thermal conductivity detector, was used for this purpose. Column specifications and operating conditions used for the fractionation and collection of the various compounds are shown in Table I (bottom). A flow rate of approximately 200 ml/min of He carrier gas was used. A fraction collector previously described by Wood and Reiser (23) was used for the collection of desired components.

Nomenclature

A shorthand system of nomenclature used to identify the isomeric glyceryl ethers has previously been described in detail (22). It is essentially a numerical system, the first number

TABLE I

Column Specifications and Operating Conditions Used for the Analysis of Glyceryl Ethers, Starting Materials and Synthetic Intermediates

Compound analyzed	% Liquid phase	Support	Column dimensions[a]	Column temp (°C)
		Analytical		
Glyceryl ether TFA deriv.	10.5 Ethylene glycol succinate methyl silicone polymer (EGSS-X)[b]	Gas-Chrom P 100-120 mesh	5 ft × ⅛ in. SS and Al	170
Glyceryl ether TFA deriv.	5 Methyl silicone polymer (SE-30)	Chromosorb W 60-80 mesh[c]	5 ft × ⅛ in. SS[d]	230
Glyceryl ether isopropylidene deriv.	16 Ethylene glycol succinate polyester (EGS)[b]	Chromosorb W 100-140 mesh	3 ft × ⅛ in. Al	220
Alkyl iodides and bromides	16 EGS	"	10 ft × ⅛ in. Cu and Al	185
n-propyl Octadecyl ether	"	"	3 ft × ⅛ in. Al	190
3-pentyl Octadecyl ether	"	"	"	200
Oleyl and linoleyl alcohol TMS ether deriv.	"	"	"	150
Oleyl- and linoleyl-p-toluene sulfonates	"	"	"	210
		Preparative		
18:1-1 TMS ether deriv.	25 SE-30[e]	Gas-Chrom Rz[b] 50-60 mesh	7 ft × ⅜ in. Al	285
n-propyl Octadecyl ether	"	"	10 ft × ⅜ in. Al	200
3-pentyl Octadecyl ether	"	"	"	265

[a] Columns were stainless steel (SS), Aluminum (Al) and Copper (Cu).

[b] Obtained from Applied Science Laboratories, State College, Pa.

[c] Acid washed support.

[d] Pretested packed columns obtained from Wilkens Instrument & Research, Inc., Walnut Creek, Calif.

[e] Liquid phase coated on support by flash evaporation technique.

represents the number of carbon atoms in the hydrocarbon chain, the second number represents the number of double bonds in the chain and the third denotes to which carbon atom of the glycerol molecule the hydrocarbon chain is bonded. The sometimes used α and β system of denoting isomers is synonymous with the 1 and 2 systems used here.

Infrared Spectroscopy

Infrared spectra were obtained with a Perkin-Elmer (Perkin-Elmer Instrument Div., Norwalk, Conn.) Model 521 Grating Infrared Spectrophotometer. Spectra of solvent-free liquids were obtained by sandwiching between two optically-ground potassium bromide (KBr) crystals. Spectra of the solid compounds, and sometimes of liquids, were obtained with 300 mg KBr discs containing approximately 1.2 mg of sample. A high vacuum cell equipped with cesium bromide windows was used to obtain a spectrum of 18:1-1 in a KBr pellet at liquid nitrogen temperature. An IR spectrum was obtained for each of the compounds from 2.5 to 40 μ (4000 to 250 cm^{-1}). Regions 4000 to 3700 cm^{-1}, 2700 to 1700 cm^{-1}, and 500 to 250 cm^{-1} of little or no absorption were not reproduced in the spectra shown.

Nuclear Magnetic Resonance Spectroscopy

A Varian (Varian Associates, Analytical Instrument Div., Palo Alto, Calif.) A-60 High Resolution Spectrometer was used to measure the spectra of the glyceryl ethers and related compounds. Unless otherwise noted, spectra were obtained with a 15% solution of each compound in carbon tetrachloride. All nuclear magnetic resonance spectra were measured at room temperature, except the 1- and 2-isomers of 14:0, and 16:0, and 18:0 glyceryl ethers, which were run at 50C. The single proton resonance peak of tetramethylsilane (TMS) used as an internal standard was assigned to value of zero parts per million (ppm) of the total magnetic field.

Thin-Layer Chromatography

Silica Gel G plates impregnated with approximately 10% sodium arsenite were prepared according to the procedure of Morris (24). Boric acid ($\sim$ 5.0%) impregnated Silica Gel G plates were prepared by the method of Thomas et al. (25). One part of Silica Gel G was slurried with two parts of water for the preparation of unimpregnated plates. Uniform 0.25mm and 1.0mm adsorbent layers were spread on 5 $\times$ 20 and 20 $\times$ 20 cm glass plates with a Colab No. 2810 applicator (Colab Laboratories, Inc., Chicago Heights, Ill.), modified in this laboratory (26). After the chromatoplates had air-dried for 30 min, they were activated in an oven for 30 min at 110C. The boric-acid-impregnated plates were activated 2 hr at the same temperature. The 1.0-mm thick preparative plates were used for the isolation and purification, while the 0.25-mm plates were used for analytical determinations. The saturated glyceryl ethers were separated from the long-chain alkyl halide by chromatography of the reaction mixture on 20- $\times$ 20-cm plates (1.0-mm thick adsorbent layer) with a hexane-diethyl ether-methanol, 80:20:5 (v/v/v) solvent system. Unsaturated glyceryl ethers were separated from the long-chain alcohols by chromatography of the reaction mixture on preparative plates developed in hexane-diethyl ether-methanol, 80:20:10 (v/v/v). The silver ion thin-layer chromatography (TLC) system recently reported (22) was used to check the glyceryl ethers for contamination by ethers of the same chain length, varying only in degree of unsaturation. Boric acid and sodium arsenite impregnated plates developed in chloroform-methanol, 98:2 (v/v) were used to check isomeric purity in addition to GLC analyses. Separations on analytical TLC plates were visualized by charring according to the procedure of Privett and Blank (27). The glyceryl ether region of the heavily loaded preparative plates was located visually, or more distinctly with the aid of UV light. The desired region was scraped from the plates and the glyceryl ethers were extracted with several volumes of diethyl ether.

Melting Point Determinations

Melting points were determined in triplicate for each compound. Samples were heated to 125C in the capillary melting point tubes and then quickly cooled in an ice bath to obtain only one polymorphic form in compounds capable of existing in more than one crystalline form.

EXPERIMENTAL

Synthesis of Saturated 1- and 2-Glyceryl Ethers

Saturated 1- and 2-glyceryl ethers ranging in hydrocarbon chain length from C_{10} to C_{18} were prepared from potassium salts of 1,2-isopropylidene glycerol and 1,3-benzylidene glycerol, respectively, by reacting with a twofold excess of alkyl iodides or bromides according

to the procedure of Davies et al. (4). The procedure was scaled down to prepare 300-600 mg of glyceryl ethers. The 1,3-benzylidene glycerol was prepared according to the procedure of Mattson and Volpenhein (28). The reaction mixtures containing the 1,2-isopropylidene glyceryl ethers or the 1,3-benzylidene glyceryl ethers were hydrolyzed, according to the procedure employed by Hartman (29) to remove these same blocking groups from monoglycerides. The glyceryl ethers were separated from the contaminating alkyl halides, hydrocarbons, etc., by preparative TLC (see TLC section). One- and 2-glyceryl ethers were further purified by crystallization from approximately 50 to 200 volumes of hexane, respectively, at –20C.

Synthesis of Unsaturated 1- and 2-Glyceryl Ethers

Mono- and diunsaturated C_{18} 1- and 2-glyceryl ethers were prepared from the potassium salts of 1,2-isopropylidene glycerol and 1.3-benzylidene glycerol, respectively, by reacting with the desired alkenyl-*p*-toluenesulfonate according to the procedure of Gupta and Kummerow (30). The alkenyl-*p*-toluenesulfonates were prepared by reacting *p*-toluenesulfonyl chloride with the unsaturated alcohols (6) and used without purification. Oleyl and linoleyl alcohols were prepared by reducing the corresponding methyl esters with lithium aluminum hydride (31). The blocking groups were hydrolyzed with boric acid. Contaminating alcohols, toluenesulfonates, etc., were separated from the saturated 1- and 2-glyceryl ethers by preparative TLC (see TLC section) and further purified by crystallization from hexane as described earlier.

Synthesis of n-Propyl and 3-Pentyl Octadecyl Ethers

Normal propyl and 3-pentyl octadecyl ethers were prepared to aid in the interpretation of the IR and NMR spectra of the isomeric glyceryl ethers. Synthesis was achieved by reacting the alkyl halide with the potassium salts of n-propyl and 3-pentyl alcohols, respectively. Approximately 0.5 g of 98% + pure (as determined by GLC) *n*-propyl octadecyl ether (mp 28.2-28.6C) and 0.5 g of 95% + pure 3-pentyl octadecyl ether (mp 13.0-14.7 C) were isolated by preparative GLC. The contaminant in each case was 1-iodooctadecane which did not interfere with the IR or NMR spectra.

Preparation of Glyceryl Ether Derivatives

Trifluoroacetate (TFA) and trimethylsilyl (TMS) ether derivatives of the isomeric glyceryl ethers were prepared according to the procedure recently described (22). The glyceryl ether diacetates were prepared by the method of O'Brien and Rouser (32). The last traces of pyridine were removed from the glyceryl ether TMS derivatives by preparative GLC.

Materials

The methyl esters used for the preparation of the alcohols were obtained from the Hormel Foundation, Austin, Minn. The 1,2-isopropylidene glycerol (2,2-dimethyl-1,3-dioxolane-4-methanol), 1-alkyl halides, *p*-toluenesulfonyl chloride, and trifluoroacetic anhydride were obtained from Eastman Organic Chemicals, Rochester, N. Y. Other materials were purchased as follows: Hexamethyldisilazane from Peninsular Chemical Research, Gainesville, Fla.; trimethylchlorosilane from K & K Laboratories, Plainview, N.Y.; and trimethyl borate from Matheson Coleman & Bell, East Rutherford, N. Y. Solvents and other reagents were reagent grade and used without further purification.

RESULTS AND DISCUSSION

Synthesis of Isomeric Glyceryl Ethers

The progress of the reactions was monitored by GLC analysis of samples removed intermittently from the reaction flask and injected directly into the chromatograph for each isomer. Using this technique we found that, on the average, overall yields of 50-60% of the 1- and 2-isomers could be obtained in 5-7 hr for both the saturated and unsaturated glyceryl ethers. Additional reaction time, up to 24 hr, usually did not increase the yield appreciably. Alkyl bromides and alkyl iodides were found to give equally high yields. Occasionally we observed substantial quantities of 1-alkenes (dehydrohalogenation products of 1-alkyl iodides) presumably caused by excess potassium.

Copper columns were found to cause an apparent decomposition of the alkyl iodides. Aluminum columns packed with the same material operating under identical conditions gave satisfactory results. It was not established whether the alkenyl-*p*-toluenesulfonates were eluted from the column intact or as a major breakdown product. The latter appeared unlikely since only one major peak was eluted for each alkenyl-*p*-toluenesulfonate without the leading front or tailing, usually associated with decomposition.

TABLE II
Melting Points and Purity of the Isomeric Glyceryl Ethers and Derivatives

Compound[a]	Melting point (°C)[b]	% Purity	Compound[a]	Melting point (°C)[b]	% Purity
10:0-1	34.5-36.0	97	18:1 (9c)-2	13-14	99+
10:0-2	30.4-31.5	97	18:2 (9c, 12c)-1	-4.5 to -4.0	95
12:0-1	47.2-48.0	99+	18:2 (9c, 12c)-2	-0.8 to 0	95+
12:0-2	45.0-46.5	99+	18:0-1 TFA	40.5-42.1	98
14:0-1	55.4-56.1 44.8-45.3[c]	98	18:0-2 TFA	25.2-26.2[c] 36.4-37.1	98
14:0-2	54.9-55.4	98	18:0-1 TMS	<20	99+
16:0-1	59.9-60.6	97	18:0-2 TMS	<20	99+
16:0-2	62.5-63.3	97	18:0-1 Diacetate	32.9-33.7	98
18:0-1	65.0-65.7	98	18:0-2 Diacetate	29.0-30.0	98
18:0-2	68.5-69.0	98+	n-propyl octadecyl	28.2-28.6	99+
18:1 (9c)-1	17.6-18.5	99+	3-pentyl octadecyl	13.9-14.7	95

[a] Shorthand system of nomenclature previously described (22).
[b] Mean range of three determinations.
[c] Polymorphic forms.

Purity of Glyceryl Ethers and Derivatives

Purity of the isomeric glyceryl ethers was determined by GLC analyses of their TFA derivatives on polar EGSS-X and nonpolar SE-30 columns (22) and by TLC on Silica Gel G layers impregnated with silver nitrate (22), sodium arsenite and boric acid, and on unimpregnated Silica Gel G. The purity for each of the ethers and derivatives prepared from them are given in Table II. All the ethers were greater than 99% pure isomerically and as a class, and more than 95% pure as a single identifiable component. The 1-isomers, of course, represented a racemic mixture. The impurities were glyceryl ethers with hydrocarbon chains two carbon atoms longer or shorter. The use of the alkenyl-*p*-toluenesulfonates produced no isomerization of double bonds detectable by IR in our synthetic procedures; the isomerization observed by Baer et al. (6) suggests that the sodium naphthalene used in their synthetic scheme caused the isomerization. Baer and Fisher (7) later found no isomerization using the tosyl-isopropylidene glycerol, oleyl alcohol, and sodium naphthalene.

The IR and NMR spectra and the melting points reported here also indicated that the glyceryl ethers and derivatives were quite pure. The melting points obtained are shown in Table II. Melting points obtained are on the average 4-5 degrees lower than those reported by Baumann and Mangold (33) for a similar homologous series of saturated 1-isomers. Melting points reported by numerous investigators (2,5,30,34-37) for the 18:0-1 and 16:0-1 ethers ranged from 68-72C and 59-64C, respectively. The 18:1-1 melting point was in close agreement with the 18-19C and 17.6-19C values reported by Baumann and Mangold (33), and Baer and Fisher (7), respectively. The 8C mp reported by Baumann and Mangold (33) for the 18:2-1 is 12 degrees higher than that reported here. The 18:0-2 melting point agreed well with that reported by Gupta and Kummerow (30), while those values reported by other investigators (4,10 and 36) are 6-8 degrees lower. The only reported melting point for the 16:0-2 agreed well with that found (4). The synthesis of the other 2-isomers: 18:1-2, 18:2-2, 14:0-2, 12:0-2, and 10:0-2 have not previously been reported.

The discrepancies in some cases between the observed melting points and values cited in the literature can be attributed to polymorphism and uncorrected melting points. The former can give rise to larger differences in melting points, as indicated with the 14:0-1 (see Table II), than if large quantities of impurities are present. Therefore, unless all the compounds are treated in a manner similar to that described in the Analytical Methods section, to obtain a known or reproducible polymorphic form, melting points are of little value. Interestingly, when a linear plot of the melting points of the saturated 1- and 2-glyceryl ethers (Table II) are plotted on the ordinate, versus the hydrocarbon chain length on the abscissa, two curves are obtained that intersect each other when the hydrocarbon chain lengths are 14 carbon atoms long. Initially, the 1-isomers have the higher melting points, but as the hydrocarbon chain increased in length, the melting points become lower than the 2-isomers.

The melting point obtained for the 18:0-1 diacetate agreed well with the 34-35C reported by Carter et al. (10); however, they reported

the 18:0-2 diacetate as a wax, but we obtained a definite melting point a few degrees below that of the 1-isomer. The 18:0-1 and 18:0-2 TMS ether derivatives were waxy and a definite melting point was not observed. Trifluoroacetate derivatives of the 18:0-1 and 18:0-2 had surprisingly high melting points, considering their GLC behavior, and two polymorphic forms were observed with the 2-isomer.

TLC of Isomeric Glyceryl Ethers

Sodium arsenite (A) and boric acid (B) impregnated Silica Gel G chromatoplates depicting the purity and resolution of 18:1-1 (1) and 18:1-2 (2) glyceryl ethers are shown in Figure 1. The 1-isomers of monoglycerides were found to migrate lower than the 2-isomers on boric acid impregnated silica gel layers by Thomas et al. (25); however, an explanation for the lower migration was not discussed. The reversal of the migration order on the two different impregnated adsorbent layers can be explained by assuming the glyceryl ether borate complex to be more polar than the uncomplexed form. The lower spots on each plate (Fig. 1) migrated to approximately the same position as both isomers on unimpregnated Silica Gel G plates, in the same solvent system. If we assume that arsenite and borate ions complex in a ratio of 1:1 with the glyceryl ethers [a ratio shown to exist by Roy et al. (38) with several polyols] and that the uncomplexed glyceryl ethers are more polar than the complexes (39), then the compounds on plate A lane 1 and plate B lane 2 represent 1,2-diol arsenite and 1,3-diol borate complexes, respectively. The latter, to the authors' knowledge, has not been reported to form a strong borate complex. Compounds on plate B (uncomplexed 2-isomers) with a higher R_f corresponds to the compounds on plate A (uncomplexed 2-isomers) with a lower R_f. Relatively higher R_f values on borate-impregnated plates compared to unimpregnated and sodium arsenite impregnated adsorbent layers developed in the same solvent system, have been reported by Morris (24). This is presumably caused by a reduction in the number of adsorption sites of the silica gel. Equations 1 and 2 are also consistent with this hypothesis.

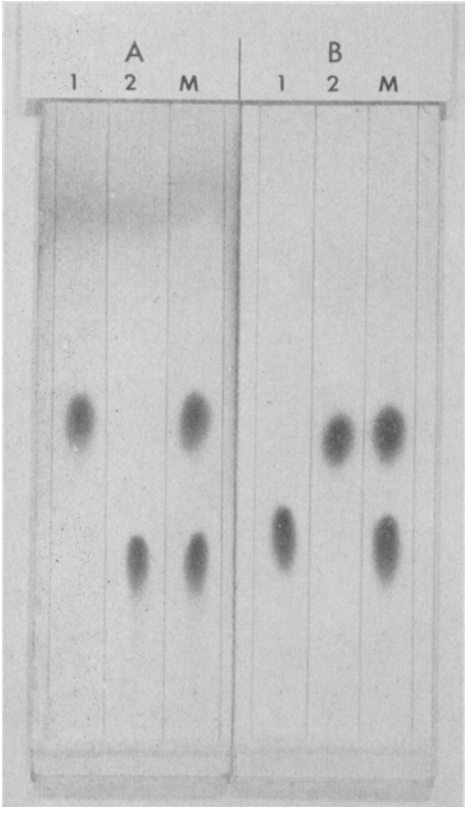
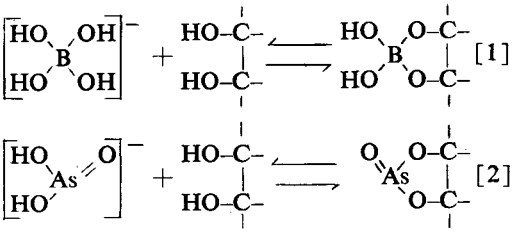


FIG. 1. TLC separation of 18:1-1 (1) and 18:1-2 (2) isomeric glyceryl ethers on Silica Gel G adsorbent layers impregnated with sodium arsenite (A) and boric acid (B).



A complex formed between a 1,2-diol and a borate ion by splitting out two molecules of water could give rise to a more polar complex than the original diol, as suggested by Roy et al. (38). This agrees with equation 1 and our TLC data. A complex formed between a 1,2-diol and a monohydrated arsenite ion by splitting out two molecules of water would be expected to be less polar than the original diol, which agrees with equation 2 and our TLC data. Such an arsenite ion could not be confirmed or excluded by the work of Roy et al. (38).

Comparison of Isomeric Glyceryl Ether IR Spectra

The IR spectra shown in Figure 2 were obtained from 13-mm diameter transparent KBr pellets. The first four spectra are included to

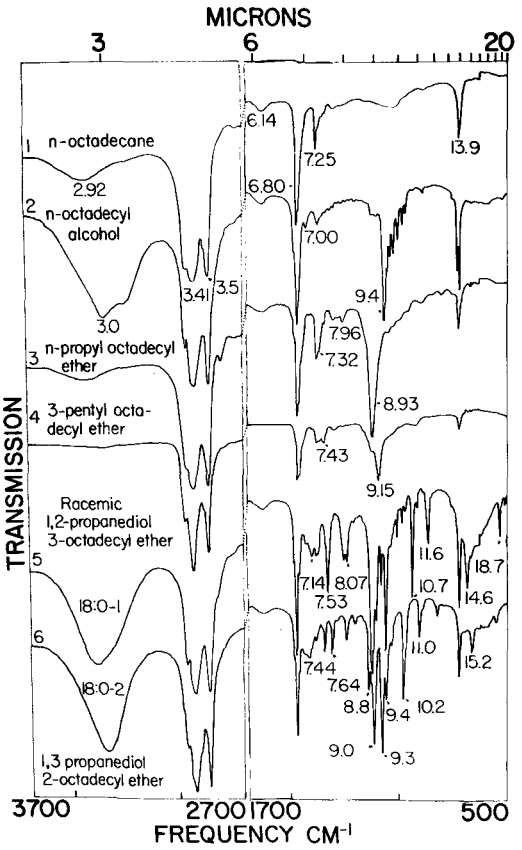


FIG. 2. IR spectra of 18:0-1 and 18:0-2 isomeric glyceryl ethers and compounds with individual functional groups found in the two isomeric ethers.

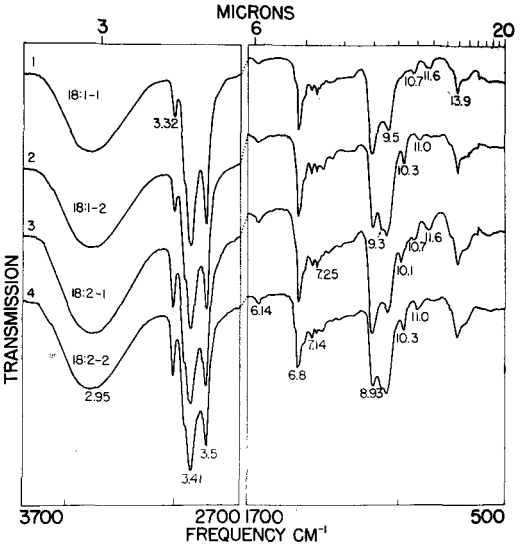


FIG. 3. Infrared spectra of long-chain synthetic mono- and diunsaturated 1- and 2-glyceryl monoethers.

aid in the interpretation of the last two isomeric glyceryl ether spectra. Absorption areas in the glyceryl ether spectra, due to the hydrocarbon portion of the molecule, are indicated by the n-octadecane spectrum, and similarly the primary alcohol, primary and secondary ether absorption areas are indicated by spectra 2, 3 and 4, respectively. The differences in absorption between primary and secondary ethers that are slightly shifted from 8.93 and 9.15 μ in the glyceryl ethers are shown by the *n*-propyl and 3-pentyl octadecyl ether spectra. As shown in Fig. 2 (5 and 6), the isomeric glyceryl ethers are easily distinguishable by other differences in absorption that are probably due to the presence of the secondary alcohol group in the 1-isomer (absent in the 2-isomer) that can exhibit intramolecular hydrogen bonding with the adjacent primary alcohol group. The spectrum of the 18:0-2 glyceryl ether was similar to that obtained by Debuch (40). The IR spectra obtained for the 10:0, 12:0, and 14:0 and 16:0 1- and 2-glyceryl ethers were similar to those of 18:0-1 and 18:0-2 (5 and 6) isomers shown in Fig. 2. The small absorption bands at 6.14 μ in all spectra and 2.92 μ in compounds 1, 3 and 4 were due to atmospheric water vapor and residual moisture in the ethers.

Spectra of the isomeric mono- and diunsaturated glyceryl ethers are shown in Figure 3. These, like the saturated 1- and 2-glyceryl ethers, are distinguishable by IR. The monounsaturates are only distinguishable from the diunsaturates by the degree of absorption in the region of 3.32 μ. Slight absorption at 10.7 and 11.6 μ in the 1-isomers differs from the weak 10.3 and 11.0 μ bands of the 2-isomers, and both isomers are further distinguishable by the stronger absorption at 9.3 and 9.5 μ of the latter. The 18:1-1 spectrum was not improved when the sample was analyzed as a KBr disc at ambient or liquid nitrogen temperatures. This was unexpected since Chapman (41) has shown that, generally, decreased resolution or increased absorption band smearing occurs with increased temperature; however, the converse relationship does not appear to hold. One possible explanation is that the dispersed glyceryl ether in the KBr disc may have little opportunity to rearrange its crystalline form.

IR Spectra of 18:0-1 and 18:0-2 Derivatives

Spectra of the 18:0-1 and 18:0-2 glyceryl ether diacetate, TFA and TMS derivatives are shown in Fig. 4. The isomeric diacetates differ by the stronger absorption in the region of 9.5 μ of the 2-isomer and the presence of the two weak absorption bands at 9.7 and 10.4 μ in the 1-isomer, absent in the 2-isomer. The contribution of the hydrocarbon chain to the spectra can be obtained by comparison with spectrum 1, Fig. 2. The slight absorption in the region of 2.9 μ is probably due to water vapor, since the 6.14 water band is also present. The ester carbonyl band at 5.7 μ, and assignments to other absorption bands have been made by Carter et al. (10).

The spectra of the two isomeric 18:0 glyceryl ether TFA derivatives are also shown in Fig. 4, and are distinctly different. However, the differences may be attributable to a different polymorphic form for each isomer or a mixture of the two observed forms of the 18:0-2 isomer. The carbonyl ester band shifted from 5.7 μ in the isomeric diacetates to 5.55 μ in the 1-isomer, and a doublet at 5.55 and 5.61 in the 2-isomer of the TFA derivatives.

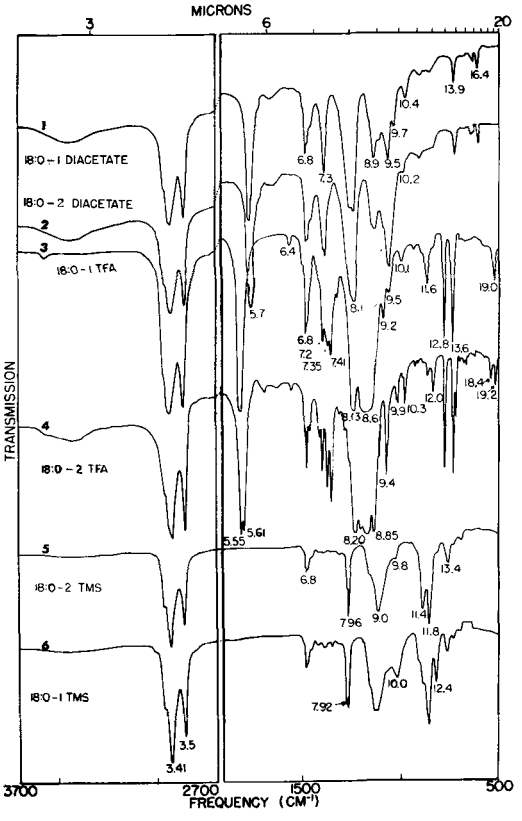


FIG. 4. Comparison of IR spectra of 18:0-1 and 18:0-2 glyceryl ether diacetate, TFA and TMS derivatives.

Spectra of the 18:0 isomeric glyceryl ether TMS ether derivatives are depicted at the bottom of Fig. 4. The broad ether absorption band in the region of 9.0 μ consisting of several observable overshadowed absorption bands, probably resulted from differences in absorption of primary and secondary silyl and alkyl ethers. The two isomers are distinguishable by the presence of two absorption bands at 10.0 and 12.4 μ in the 1-isomer, absent in the 2-isomer, and by the differences in degree of absorption at 11.4 and 11.8 μ of the two isomers.

Normal Propyl and 3-Pentyl Octadecyl Ether NMR Spectra

Normal propyl and 3-pentyl octadecyl ethers were synthesized to aid in the interpretation of the isomeric glyceryl ether spectra. The single hydrogen on carbon number 3 of the 3-pentyl octadecyl ether gave rise to a quintuplet at 3.02 ppm (central peak), which is partially overlapped by a triplet (central peak 3.32 ppm) arising from the two protons of the hydrocarbon chain adjacent to the ether oxygen. The resonance of the protons adjacent to the ether oxygen in the *n*-propyl octadecyl ether appear as two practically superimposable triplets at 3.33 ppm (central peak). The NMR spectrum of *n*-propyl hexadecyl or octadecyl ether has previously been obtained by Carter et al. (10); however, resolution was not sufficient to observe the two triplet sets for the methylene protons adjacent to the ether oxygen.

NMR Spectra of Isomeric Glyceryl Ethers

The NMR spectra of mono- and diunsaturated isomeric pairs, 18:1 and 18:2, and of two saturated glyceryl ethers, 12:0-1 and 16:0-2 are shown in Fig. 5. Spectra of the 1- and 2-isomers of all the glyceryl ethers analyzed were readily distinguishable and distinctly different. The partially visible broad-line resonance at approximately 3.75 ppm, equivalent to one proton, observed in the 1-isomers (absent in the 2-isomers) was assumed to be due to a downfield shift of the lone hydrogen on carbon atom number 2 of the glycerol moiety caused by the deshielding effect of the electronegative environment. Resonance of this same proton in the 2-isomers occurred upfield at approximately 3.42 ppm, which may or may not represent the central peak of a partially visible

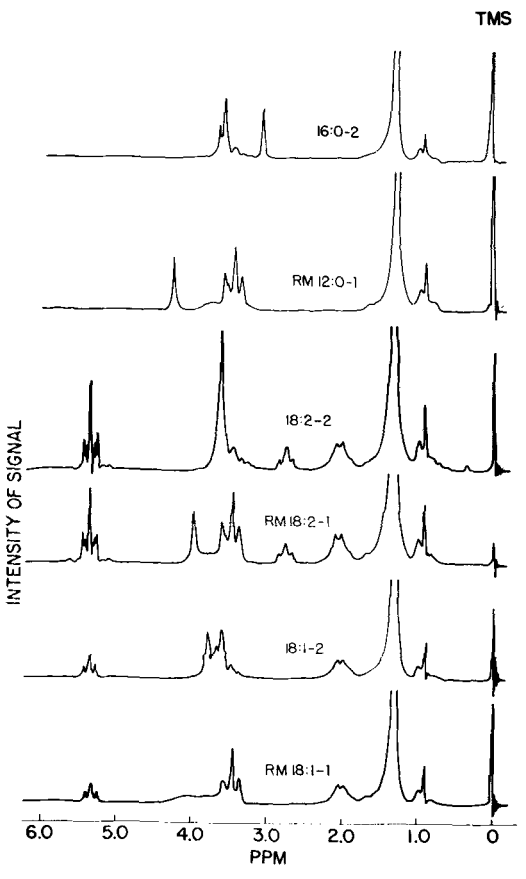


FIG. 5. Proton resonance spectra of saturated, mono-, and diunsaturated isomeric glyceryl ethers obtained at 60 mc.

multuplet (when not obscured by hydroxyl proton resonance).

The unresolved resonance at 3.65, 3.58, and 3.53 ppm of the 2-isomers differs markedly from the resonance shift upfield to 3.58, 3.53, 3.42, and 3.32 ppm in the 1-isomers. Resonance at 3.65 ppm in the 2-isomers, absent in the 1-isomers, is probably due to the methylene protons on carbon atoms 1 and 3 of the glycerol moiety. The spectra of the 2-isomers are further complicated by the triplet representing the methylene protons of the hydrocarbon chain adjacent to the ether oxygen. The spectra of the 1-isomers in the area of 3.3-3.7 ppm are equally complicated, and resonance-band assignments, other than relative expected order, are not made. The 3-glyceryl methylene hydrogens of the 1-isomers, like those of the 2-isomers, are expected to appear downfield, followed closely by a triplet and two doublets for the methylene hydrogens of the hydrocarbon chain and the nonequivalent methylene hydrogens on carbon number 1 of the glycerol moiety. Although specific assignments for this portion of the molecule were not possible, the usefulness of the NMR spectra is not limited.

The hydroxyl proton resonance, which is known to exhibit marked dependence on concentration and temperature (42) is shown in Table III for the isomeric glyceryl ethers. The hydroxyl proton resonance of the 1-isomers occurs at a lower field than the 2-isomers. Increased concentration of the 1-isomers also resulted in resonance of the hydroxyl protons at lower field. The broad resonance line, exhibited only by the 18:1-1 at two concentrations, is not understood.

Both isomers of the monounsaturated glyceryl ethers are easily distinguished from the diunsaturated by the resonance of the methylene hydrogens between the two double bonds at 2.72 ppm and by the intensity of the signal of the vinyl hydrogens at 5.31 ppm, as shown in Figure 5. In the unsaturated glyceryl ethers the hydrogens that are allyl to the double bond, which usually are obscured by methylene hydrogens adjacent to a carbonyl ester, are observed without interference at 2.04 ppm.

NMR Spectra of Glyceryl Ether Derivatives

The spectra of the 18:1-1 diacetate (top) and TMS ether (bottom) derivatives, obtained neat at room temperature, are shown in Figure 6. The purity and origin of the 18:1-1 glyceryl

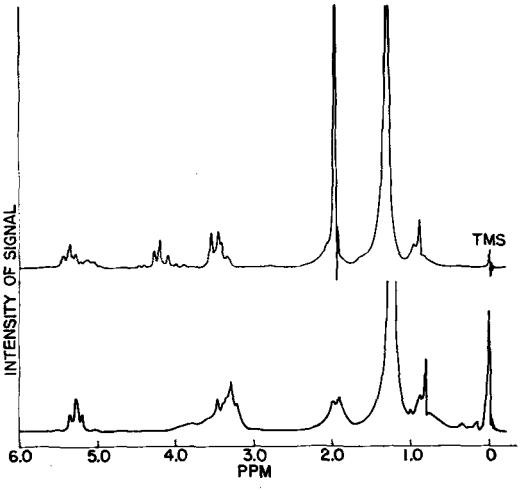


FIG. 6. NMR spectra of 18:1-1 glyceryl ether diacetate (top) and trimethylsilyl ether (bottom) derivatives obtained at 60 mc.

TABLE III
Chemical Shifts of Isomeric Glyceryl Monether Hydroxy Protons

Glyceryl ether	OH chemical shift (ppm)	Conc (%)	Temp
12:0-1	4.25	25	Room
12:0-2	3.43	10	Room
14:0-1	3.48	15	50C
14:0-2	2.92	15	50C
16:0-1	3.33	15	50C
16:0-2	3.08	15	50C
18:0-1	3.30	15	50C
18:0-2	2.92	15	50C
18:1-1	4.04[a]	15	Room
18:1-1[b]	4.48	Neat	Room
18:1-2	3.76	15	Room
18:2-1	3.94	15	Room
18:2-2	3.60	15	Room

[a] Broad band, see Fig. 6.

[b] Commercially available sample from Western Chemical Ind. Ltd., Vancouver, Canada, greater than 95% pure as a class (sterols main contaminant) and approximately 90% monoene 1-isomers.

ether is given at the bottom of Table III. The broad resonance band of the lone 2-glyceryl proton appears downfield at approximately 5.15 ppm in the diacetate and remains at 3.75 ppm in the TMS ether, identical to the free glyceryl ethers (Fig. 5). The two nonequivalent 3-glyceryl methylene hydrogens resonate at 4.18 ppm (central peak) as two partially overlapped doublets in the diacetate, and, as expected, these hydrogens appear unresolved from the other protons adjacent to the ether oxygens at 3.28 ppm (central peak) for the TMS ether. Resonance in the diacetate at 3.40 (central peak of triplet) and 3.50 ppm (center of doublet) arises from the hydrogens adjacent to the ether oxygen. The two doublets of the nonequivalent 1-glyceryl methylene hydrogens were superimposed. Aside from the acetate methyl proton resonance at 1.95 ppm, all resonance bands for the two derivatives are the same as previously discussed. The NMR spectra of the 18:0-1 and 18:0-2 glyceryl ethers and monoglyceride diacetates have previously been obtained at 40 mc by Carter et al. (10), and assignments have been made. Resolution was poor, but sufficient to demonstrate the 2-glyceryl ether diacetate (not determined here) spectrum distinguishable from that of the 1-isomer.

The IR and NMR spectra of the long-chain isomeric 1- and 2-glyceryl monoethers and derivatives, both saturated and unsaturated, have served to demonstrate the applicability of these two types of spectroscopy for the characterization and structural determination, in addition to distinguishing between the two isomeric forms.

ADDENDUM

While this manuscript was being reviewed for publication, a paper by Serdarevich and Carroll (43) appeared dealing with the physical and chemical properties of two isomeric pairs of anteiso glyceryl ethers and the 16:0 isomers. Their findings are compatible with those reported here.

ACKNOWLEDGMENT

NMR and IR spectra obtained by Miss Lucy Scroogie and P. E. Staats, Oak Ridge National Laboratory (ORNL).

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[Received May 6, 1966]