All retinol derivatives saturated at the terminal oxygen (methyl retinyl ether, retinol, retinyl acetate, and retinyl palmitate) tended to form anhydro retinol upon exposure to hot columns and metal tubing. A free radical–induced dehydration of this group of compounds is favored over the well known acid catalyzed reaction on the following grounds: Injection of NH_3 or pyridine, which should neutralize acidic sites, did not decrease the dehydration. The relative rates of dehydration in acid media are retinol > methyl retinyl ether > retinyl acetate (*3*). During gas chromatography, the relative rates are essentially inverted—e.g., retinyl acetate > retinol > methyl retinyl ether. Hydroquinone, a free radical inhibitor, decreased dehydration in columns packed with glass beads.

Further study of the mechanism by which β-carotene and hydroquinone inhibit the dehydration reactions would be welcome and might lead to the introduction of other suitable protective agents for retinol and other unstable compounds.

The dehydration reaction has been used in liquid medium as an assay procedure for free retinol (*4*). Similarly, the formation of anhydro retinol during gas chromatography could be used as an assay for retinol, its ethers, and its esters if the procedure were carried out under conditions conducive to dehydration—i.e., high column temperatures and long retention times. This assay might be particularly applicable to the natural esters of retinol, which cannot be chromatographed without destruction at this time.

LITERATURE CITED

(1) Commission on Nomenclature of Biological Chemistry, International Union of Pure and Applied Chemistry, *J. Am. Chem. Soc.* **82**, 5575 (1960).
(2) Farrar, K. R., Hamlet, J. C., Henbest, H. B., Jones, E. R. H., *J. Chem. Soc.* **1952**, p. 2657.
(3) Hanze, A. R., Conger, T. W., Wise, E. C., Weisblat, D. I., *J. Am. Chem. Soc.* **70**, 1253 (1948).
(4) Harashima, K., Okazaki, H., Aoki, H., *J. Vitaminol.* **7**, 150 (1961).
(5) Moore, T., "Vitamin A," pp. 43–66, Elsevier, Amsterdam, 1957.
(6) Ninomiya, T., Kidokoro, K., Horiguchi, M., Higosaki, N., *Bitamin* **27**, 349 (1963).
(7) Robeson, C. D., Cawley, J. D., Weisler, L., Stern, M. H., Eddinger, C. C., Chechak, A. J., *J. Am. Chem. Soc.* **77**, 4111 (1955).
(8) Shantz, E. M., Cawley, J. D., Norris, D. E., *Ibid.*, **65**, 901 (1943).

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Use of a Mass Spectrometer as a Detector and Analyzer for Effluents Emerging from High Temperature Gas Liquid Chromatography Columns

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▶ **A modified Atlas CH4 mass spectrometer has been coupled to a gas liquid chromatography (GLC) column. As the compounds emerge from the column they are ionized in the ion source of the mass spectrometer and about 10% of the total ion current is used for continuous registration of the effluent. The temperature of the column can be regulated from 50° to 350° C. by using the temperature programmer. Two molecule separators are coupled in series between the column and the gas inlet line of the mass spectrometer. With this technique the sample-to-helium ratio is increased at least 100 times. Less than 1 μg. of material introduced onto the column suffices for a good mass spectrum. The mass range *m/e* 12 to 500 can be scanned and recorded in 1 or 2 seconds. Examples are given of the separation and mass spectrometric identification of 27 components from 200 μg. of methylated fatty acids from butter fat and the separation of a mixture of C_{19} to C_{30} hydrocarbons.**

ONLY A FEW papers have been published which describe compound instruments in which a mass spectrometer connected directly to a gas chromatograph has been used for the continuous registration of components from separated organic compounds. The separation and identification of mixtures of organic compounds using a combination gas chromatograph and time-of-flight mass spectrometer of the Bendix type was performed for the first time a few years ago (*5*, *7*). Details were given for acetone and some benzene derivatives and for some halogenated hydrocarbons. Another type of instrument, the analytical mass spectrometer Model 21–103 B (Consolidated Electrodynamics Corp.), has been used with a capillary column for the separation of the known hydrocarbons up to C_{11} (*11*). The cycloidal focusing mass spectrometer, CEC Model 21–620, has been used for the same purpose and a mixture of 16 C_9 hydrocarbon isomers were identified from the mass spectra (*4*).

For a similar application, an Atlas CH4 mass spectrometer has been connected to a capillary column (*3*).

A thermionic ionization gauge in combination with a mass analyzer system has been used as a quantitative and qualitative (QQ) detector for a gas chromatograph (*18*, *19*). This type of detector is very sensitive but not suitable for qualitative work with compounds having molecular weights higher than 50, because of insufficient mass resolution.

The above-mentioned compound instruments have usually been used for the analysis of low molecular weight compounds. In studies of compounds with higher molecular weight, techniques whereby samples collected from a gas chromatograph could be subsequently analyzed in the mass spectrometer have been used for several years (*13*). Usually the samples are collected in a cold trap. This system is time consuming and suffers from many disadvantages particularly those which are associated with the collection, handling, and introduction of the sample into the mass spectrometer. In an attempt to overcome these difficulties, work was started 4 years ago to connect a mass spectrometer with the output of a gas chromatograph. To make possible the analysis of most types of compounds that can presently be separated by gas chromatography it is necessary to have a high-mass instrument with a relatively high ion-current intensity—for a mass unit resolution at *m/e* 600. An Atlas CH4 mass spectrometer equipped with an electron multiplier and a magnet current regulator with the possibility of scanning the magnetic field to cover the mass range *m/e* 12 to 400 in 3 seconds

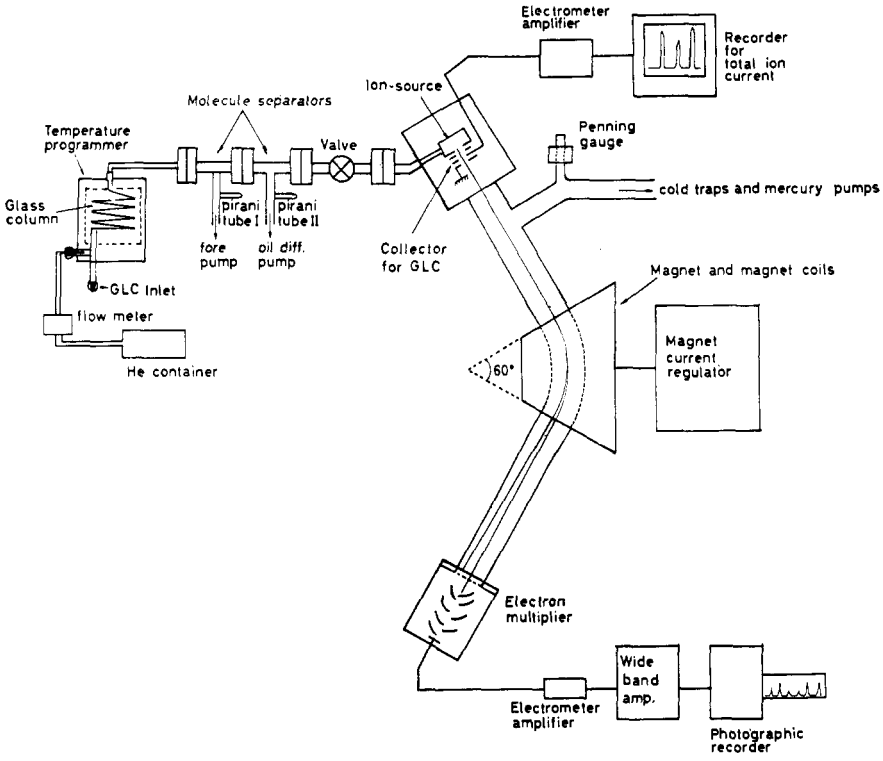

Figure 1. Schematic diagram of gas chromatograph–mass spectrometer combination

was used. A brief description of the modification of this mass spectrometer so that it can be used for fast recording has been given (*12*).

Preliminary results were obtained with methyl esters of fatty acids separated on packed columns with helium as carrier gas (*14*). These results showed that the way in which the surplus gas coming from the gas chromatography column was pumped away was very important. The construction described in the present paper was found to be the most suitable one so far and the combined gas chromatograph–mass spectrometer has proved to be reliable in daily routine work. Studies of monoterpenes using this technique have recently been published (*17*).

EXPERIMENTAL

Apparatus. Figure 1 shows the arrangement used for connecting the gas liquid chromatography (GLC) column to the mass spectrometer. Packed coiled glass columns as described by Haathi (*8*) were used. Columns with various cross sections were tested and a gas flow rate of about 2.5 ml. per sq. mm. per minute was used.

Since the gas flow into the mass spectrometer is limited to 0.1 to 0.2 ml. per minute, much of the carrier gas must be pumped away after it has passed the column and before it enters the ion source. By reducing the column diameter, the ratio of sample-to-carrier gas is increased, and less helium has to be pumped away provided the same amount of sample is introduced. When small samples are introduced onto the column, it is desirable that most of the sample should enter the ionization chamber of the mass spectrometer if one is to get a mass spectrum of good quality. This problem has been solved by feeding the effluent from the column to a molecule separator built on the principles of Becker (*1*). In this, the gas stream of the heavier molecules passes straight on through two holes of small diameter which are very close together, while the helium gas diffuses to the sides of the separator and is pumped away. The principle of using the molecule separator described by Becker was suggested by Stenhagen and Ställberg-Stenhagen has constructed and tested this type of a separator and found a 50-fold increase in the ratio of sample to helium indicated on a Philips gauge, Type PHG-010.

The molecule separators used in this work were independently constructed in this laboratory according to the principles given by Becker (*1*).

To increase the separation effect of carrier gas and compound, two molecule separators were connected in series. The line from the separators to the ion source passes an open vacuum valve. This valve was closed when the mass spectrometer was used as a separate instrument. The molecule separators are connected to two separate pumping systems, the first one to a fore vacuum pump of 6 cubic meters per hour at atmospheric pressure and the second one to an oil diffusion pump of 80 liters per second at 10^{-6} mm. Hg. The vacuum output for these separators is measured with an Autovac Pirani-type vacuum gauge made by LKB Produkter, Stockholm.

To get the line introducing the gas as short as possible, it was necessary to make a hole in the housing cup of the ion source and arrange for the line to enter directly to the ion source. A part of the total ion current was collected on a plate in the vicinity of the exit slit of the ion source. The ion current was measured with a DC electrometer amplifier and registered with a Speedomax pen and ink recorder (Type G, 0.3 second).

The Atlas CH4 mass spectrometer has been equipped with a fast recording system for scanning the mass range m/e 12 to 500 in 1 to 2 seconds. Since the last description of the mass spectrometer (*12*) a new recorder, Ultralet, manufactured by AB Elektrisk Malmletning, Stockholm, has been connected to the instrument. Since the ion-current output noise from the wide-band d.c. amplifier depends on the time constant of the ion-current measuring system, the noise was limited by using three galvanometers of 1500 c.p.s. or three of 400 c.p.s., which can be fed alternatively to the recorder by using a switch.

GLC Columns. The handling and construction of a gas chromatography column when used in combination with a mass spectrometer differs slightly from the normal procedure. It is possible to connect any gas chromatographic column to the mass spectrometer, but because of the programs that were to be investigated only packed columns have been used. The bleeding from these columns is much more critical when a mass spectrometer is used as a detector, as it will give a high background for the mass spectra. When the temperature of a 1% SE-30 column was higher than 300° C. its vapor pressure gave rise to a silicon-containing fragment which could disturb the cracking patterns of the mass spectra of the compounds to be studied. Consequently temperatures in excess of 300° C. were avoided and for higher per cent SE-30 columns the temperature must be reduced. A mass spectrum of a compound which typically appears in silicone greases is given by Biemann (*2*).

For the example given in this paper, a 4-meter × 2-mm. coiled glass column with a diameter of 10 cm. and filled with a packing of 1% SE-30 on 100- to 120-mesh acid-washed and silanized Gas-Chrom P was used (*10*) (this packing material was kindly supplied by E. C. Horning, Baylor University, Houston, Texas, and was prepared in his laboratory).

Procedure. To obtain mass spectra the acceleration voltage was kept constant at 3000 volts and the energy of electrons at 20 e.v. The same starting push-contact is used for the recording oscillograph and for the magnet current-scanning circuit, but the magnet circuit was delayed by a few tenths of a second until the photographic paper has reached a constant speed. The sample is introduced onto the gas chromatography column in the usual way.

The helium pressure at the sample-

inlet side was 1.6 kg. per sq. cm., giving helium flow of about 8 ml. per minute at the output. The temperature programming unit used was made by the F & M Scientific Corp., Wilmington, Del. Redistilled acetone or diethyl ether were used as solvents and the sample was introduced in 0.5 to 2 μl. of solvent.

To prevent large quantities of the solvent from entering the mass spectrometer analyzer tube, the vacuum valve nearest the second separator (Figure 1) was nearly closed during the time needed for the solvent to emerge from the column and be pumped out through the separators.

Part of the solvent was allowed to pass through the valve so that it would give rise to a peak on the potentiometer recorder used for registration of the total ion current. As soon as the solvent had emerged from the column, the vacuum valve was fully opened and the instrument was ready for operation as a gas chromatograph detector and for recording the mass spectra of the compounds as they emerged from the column.

As soon as the separated components coming from the column entered the ion source they were ionized and a part (approximately 10%) of the total ion current was registered on the potentiometric recorder which serves as the detector for the GLC column. When the total ion current showed a maximum value, the concentration of a sample in the ion source was at maximum. At this time the oscillograph recorder and the magnet current regulator are started for running of a mass spectrum. The mass range required is recorded and the scan can be repeated immediately. Since the scanning time is short the partial pressure normally is so constant that the relative ion-intensity variation in any given mass spectrum does not change enough to disturb the cracking pattern of the compounds studied.

RESULTS AND DISCUSSION

To obtain an accurate indication of the time when the separate components enter the mass spectrometer, a part of the total ionization current is used for the continuous registration of the gas emerging from the column. This record serves as the gas chromatograph record that enables the mass spectrometer operator to decide when to take a mass spectrum. If the carrier gas is ionized the total ion current will be very dependent upon the pressure in the ion source, resulting in a zero-line variation on the potentiometer recorder. To reduce this zero-line effect, and to prevent overloading of the d.c. amplifier, the energy of the electrons used for bombardment was set at 20 e.v. Since the ionization potential for helium is 24.8 e.v., the ion current for m/e 4 was nearly absent. The fragmentation of the organic compounds may be somewhat different if this low electron energy is used instead of the 70 e.v. normally applied, but, in all cases

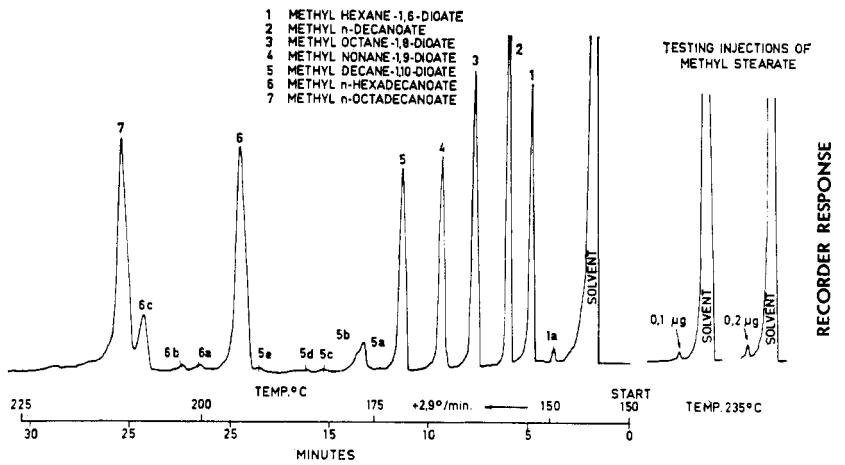

Figure 2. Chromatographic separation of mixture of fatty acid methyl esters

Glass column, 4 meters × 2 mm., containing 1% SE-30 on 100–120 mesh silanized Gas Chrom P. Initial helium flow ~8 ml./minute

studied, the general cracking pattern was the same. Table I shows the variation in intensity of some typical ion fragments from the mass spectra of methyl hexane-1,6-dioate when the energy of the electrons used for bombardment was varied from 70 to 9 e.v. At 20 e.v. it is only the smaller hydrocarbon fragments, which are considerably reduced in intensity, and the larger fragments which are the most useful ones for identification, are only slightly reduced. This table also shows the measurement of the intensity of the total ion current.

The efficiency of the molecular separators was tested by running samples with and without the separators. In both cases the pressure in the mass spectrometer tube was 5×10^{-6} mm. Hg. When the separators were not used the pressure was regulated to this value by reducing the aperture of the valve in the introducing line slightly. Samples of 5 μg. in 0.5 μl. of solvent were introduced onto the column. It was found that the total ion current was increased at least 100 times when the separators were used.

Figure 2 shows the gas chromatogram of an equimolar mixture of seven methyl esters of saturated normal, mono-, and dicarboxylic acids. The sample, 50 μg., was injected in 0.5 μl. of solvent. The temperature programmer was started 4 minutes after the introduction of the solution. The chromatogram shows that all seven compounds are well separated. However, impurities present in low concentrations are found. Sixteen mass spectra were recorded, and it was possible to determine the structure of all the observed impurities. The contaminants in the mixture were identified as methyl esters of normal saturated, normal unsaturated, and branched chain fatty acids, and one dibasic acid. Table II shows the impurities which were identified from their mass spectra. The smallest peak recorded was 5*d* (Figure 2). The amount of this compound was less than 0.1 μg. The mass spectrum shown in Figure 3 is very clear and shows a

Table I. Characteristic Ion Fragments of Methyl Hexane-1,6-dioate Obtained from Mass Spectra Taken at Different Electron Energies

	Per cent of total ionization current								
m/e	70 e.v.	50 e.v.	25 e.v.	20 e.v.	15 e.v.	12 e.v.	11 e.v.	10 e.v.	9 e.v.
15	1.4	1.2	0.5	0.2	...	...	...	...	...
27	1.2	1.1	0.2	0.1	...	...	...	...	...
41	2.4	2.3	1.1	0.6	0.4	...	...	...	...
55	7.7	8.0	5.8	4.0	2.3	0.4	...	...	...
59	10.7	11.6	10.9	9.9	8.4	4.3	2.3	1.3	...
73	3.6	3.6	3.4	2.9	1.5	0.2	...	...	...
74	5.3	5.4	6.4	6.7	6.8	5.3	5.3	3.8	...
83	3.2	3.3	3.5	3.6	3.1	2.3	3.0	2.9	...
87	1.7	1.8	1.9	1.9	1.9	1.1	0.8	0.6	...
101	6.5	7.2	9.1	11.1	14.1	22.8	27.8	30.0	36.4
111	7.7	8.1	10.1	11.1	11.4	8.5	5.3	1.7	...
114	7.7	8.3	10.6	13.0	16.0	24.91	27.8	32.1	63.6
141	0.9	0.9	1.1	1.3	1.5	3.0	4.5	7.1	...
142	4.3	4.7	5.3	6.3	7.6	11.3	10.5	5.8	...
174	0.2	0.2	0.2	0.2	0.4	1.1	1.5	1.7	...
	57100[a]	62900[a]	59600[a]	45850[a]	25800[a]	4680[a]	1330[a]	240[a]	11[a]

[a] Total galvanometer deflection in mm. of the ion beams for the mass range m/e 12–175.

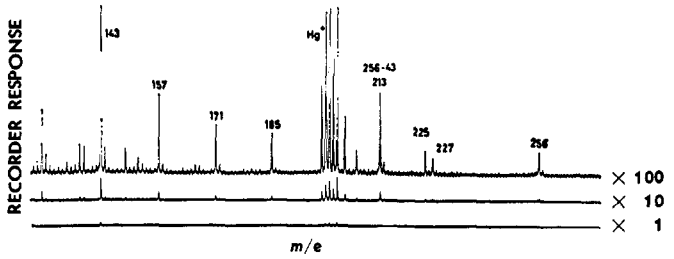

Figure 3. High-mass end of mass spectrum of component 5d (Figure 2)

Compound was identified as methyl n-methylpentadecanoate

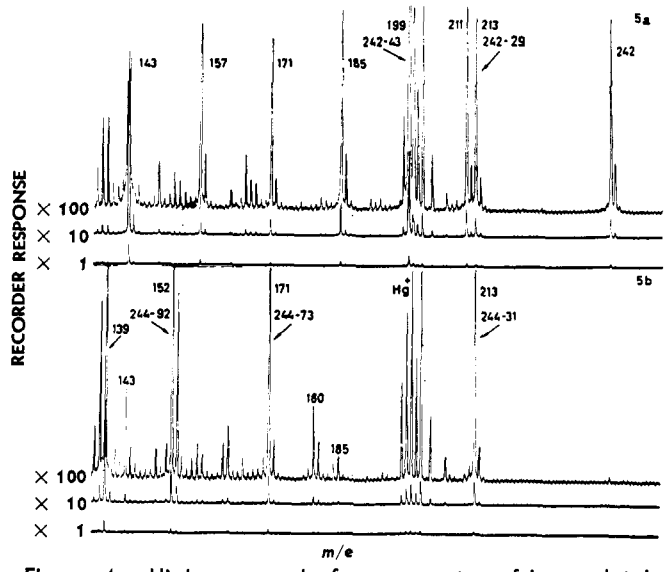

Figure 4. High-mass end of mass spectra of incompletely separated components 5a and 5b (Figure 2)

Compounds were identified as methyl n-tetradecanote and methyl undecane-1,11-dioate

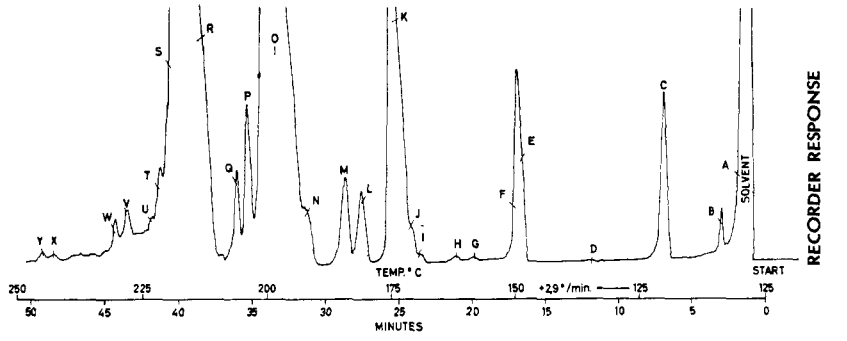

Figure 5. Chromatographic separation of methyl esters of fatty acids from butterfat

Column conditions were same as in Figure 2

parent ion at *m/e* 256 and a cracking pattern similar to that of methyl-*n*-pentadecanoate (*15*). For comparison, the same diagram (Figure 2) shows 0.1 and 0.2 μg. of methyl stearate run under the same conditions as the mixture, except that the temperature of the column in this case was kept constant at 235° C.

Figure 4 shows the high-mass end of the mass spectra of the incompletely separated components 5*a* and 5*b*. The mass spectrum of component 5*a* shows a molecular ion at *m/e* 242 and other characteristic peaks which are similar to those of methyl *n*-tetradecanoate (*15*). In the mass spectrum of the component 5*b*, no molecular ion is seen, but from peaks such as *m/e* 211 (244–31), 184 (244–60), and *m/e* 170 (244–74) the compound could be identified as methyl undecane-1,11-dioate.

Information concerning the number of double bonds, but not their position, can be obtained from the mass spectrum. It was known from earlier experiments that unsaturated compounds with one double bond and with the same number of carbon atoms in the chain gave identical mass spectra, unless the double bond was in the α-β position. It is also difficult to differentiate between a normal and an iso compound (*16*).

To illustrate the usefulness of the instruments for the separation of a naturally occurring complex mixture of fatty acids, the methyl esters prepared from butter were used. Commercial butter, 1 gram, was refluxed with 30 ml. of 10 % methanolic potassium hydroxide for 1 hour. After dilution with water the nonsaponifiable matter was extracted with petroleum ether. The water phase was acidified with hydrochloric acid and extracted twice with ether. The combined ether extracts were washed with water until neutral and evaporated to dryness. Ten milligrams of the residue was methylated with diazomethane and dissolved in 0.1 ml. of acetone, and amounts ranging from 0.5 to 2.0 μl. (50 to 200 μg. of the residue) were introduced onto the column. When a 2-μl. sample was used, several components could be observed and identified.

Figure 5 shows the gas chromatogram of the methyl esters of the residue from the butterfat prepared in this manner. A mass spectrum was taken for each component at the time which corresponds to the marks in the figure. A mass spectrum was also taken at the end of the solvent peak and it was observed that at least one component overlapped with the solvent and could be identified. It is usually advisable to check the solvent peak, to be sure that no components are missed. The smallest component recorded is shown as peak *D*, and the estimated quantity was 0.1 μg. From the mass spectra it was possible to identify a homologous series of normal saturated, normal un-

Table II. Impurities in Mixture of Mono- and Dibasic Methyl Esters of Fatty Acid Compounds

Identification based on their mass spectra only

Component	Identified compound	Mol. wt.	Quantity (est.), μg.
1*a*	Methyl *n*-octadecanoate	158	0.2
5*a*	Methyl *n*-tetradecanoate	242	0.8
5*b*	Methyl undecane-1,11-dioate	244	0.5
5*c*	Methyl 12-methyltetradecanoate	256	0.1
5*d*	Methyl *n*-pentadecanoate	256	0.1
5*e*	Methyl hexadecenoate	268	0.2
6*a*	Methyl 14-methylhexadecanoate	284	0.3
6*b*	Methyl *n*-heptadecanoate	284	0.3
6*c*	Methyl octadecenoate	296	2.0

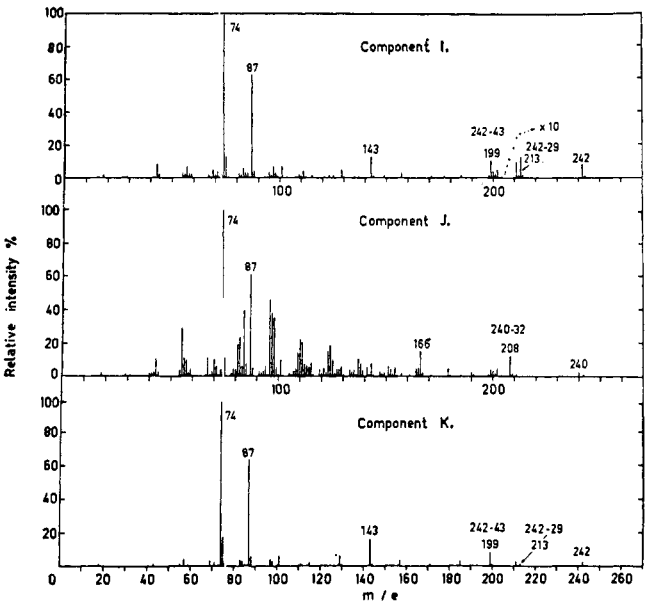

Figure 6. Mass spectra of incompletely separated components *I*, *J*, and *K* from chromatogram of butterfat (Figure 5)

Compounds were identified as methyl 11-methyltridecanoate, methyl tetradecenoate, and methyl tetradecanoate, respectively

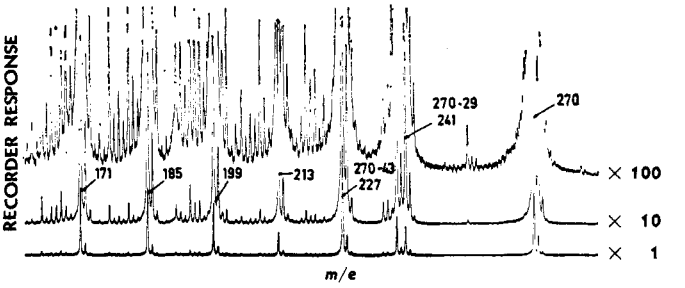

Figure 7. High-mass end of mass spectrum of component O from chromatogram of butterfat (Figure 5)

Compound was identified as methyl palmitate

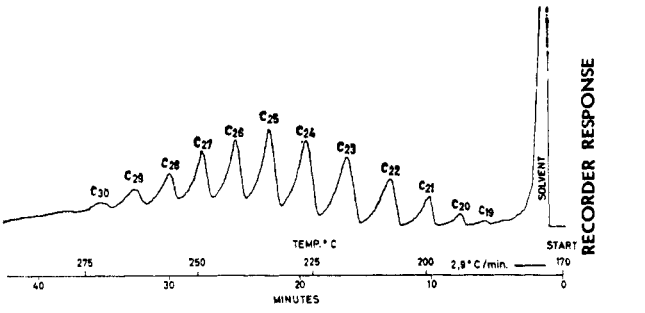

Figure 8. Chromatographic separation of typical commercial solid paraffin

Column conditions were same as in Figure 2

saturated, and branched chain fatty acid methyl esters present in this mixture. Table III lists the compounds identified from butterfat. The normal saturated methyl esters of fattyacid compounds have a chain length of 6 to 22 carbon atoms and are easy to identify (*15*). The series of homologous unsaturated compounds are observed from methyltetradecanoate to methyl docosanoate. Compounds with two and three double bonds are also identified. The mass spectra also show that several ante-iso esters are present. The presence of this type of compound in butter- and milkfat has been demonstrated by Shorland and coworkers (*6*, *9*).

Figure 6 shows the complete mass spectra of the incompletely separated components *I*, *J*, and *K* which were identified as methyl 11-methyltridecanoate, methyl tetradecenoate, and methyl *n*-tetradecanoate. Although the components overlapped, the mass spectra were distinctly different and permitted positive identification to be made. These types of compounds (*I*,*J*,*K*) seem to occur along with each of the normal fatty acid methyl esters from butterfat. Similar groups of compounds were found at both higher and lower molecular weights, and it is possible that they existed for the whole series, from methyl *n*-hexadecanoate to methyl *n*-decosanoate, but in some cases their concentrations were too low to be observed.

Figure 7 shows the high-mass end of the mass spectrum of component *O* from the chromatogram of butterfat. This component shows a parent ion at *m/e* 270 and was identified as methyl palmitate (*15*).

Figure 8 shows a gas chromatogram of a typical solid paraffin (Shell, Indonesia). Fifty micrograms of the paraffin dissolved in 1 μl. of acetone were introduced onto the column. Twelve components were observed and mass spectra were taken for each one. From these spectra it was possible to positively identify this sample as a series of normal hydrocarbons containing 19 to 30 carbon atoms.

Figure 9 shows the high-mass end of the mass spectra of the components C_{24} and C_{25} with molecular ion peaks at *m/e* 338 and 352. Contribution from the nearest lower homologs occur for both mass spectra. These mass spectra are identical with No. 575 and 1313 from mass spectral data published by the American Petroleum Institute.

The separations described merely serve as examples of the usefulness of the instrument. Several other classes of organic compounds such as diterpenes, ketones, alcohols, ketonic steroids, and sterols have been analyzed, with good results and these experiences have shown that all compounds which can be subjected to gas chromatography may also be analyzed with the compound instrument.

A number of advantages with an instrument of this type are evident: A compound to be analyzed does not require extensive purification and, if it is desired, the contaminants can be separately identified by their mass

Table III. Identified Methyl Esters of Fatty Acids from Butter Fat

Identifications based on their mass spectra only

Component	Identified compound	Mol. wt.
A	Methyl *n*-hexanoate	130
B	Methyl *n*-octanoate	158
C	Methyl *n*-decanoate	186
D	Methyl *n*-undecanoate	200
E = *F*	Methyl *n*-dodecanoate	214
G	Methyl 10-methyldodecanoate	228
H	Methyl *n*-tridecanoate	228
I	Methyl 11-methyltridecanoate	242
J	Methyl tetradecenoate	240
K	Methyl *n*-tetradecanoate	242
L	Methyl 12-methyltetradecanoate	256
M	Methyl *n*-pentadecanoate	256
N	Methyl hexadecenoate	268
O	Methyl *n*-hexadecanoate	270
P_1	Methyl 14-methylhexadecanoate	284
P_2	Methyl heptadecenoate	282
Q	Methyl *n*-heptadecanoate	284
R_1	Methyl octadecandienoate	294
R_2	Methyl octadecenoate	296
S	Methyl *n*-octadecanoate	298
T_1	Methyl 16-methyloctadecanoate	312
T_2	Methyl nonadecenoate	310
U	Methyl *n*-nonadecanoate	312
V	Methyl eicosenoate	324
W	Methyl *n*-eicosanoate	326
X	Methyl docosenoate	352
Y	Methyl *n*-docosanoate	354

spectra. Components which are poorly resolved on the GLC column can be identified since a mass spectrum can be taken every 1 or 2 seconds and observations of mass spectra can be made almost continuously during the time the effluent passes the ion source. The number of samples that the mass spectrometer can analyze in a day is increased since it is not necessary to break the vacuum to introduce a sample. Solids, liquids, and gases can all be introduced onto the mass spectrometer through the same gas chromatography inlet.

The high accuracy and sensitivity and the short time required for the analysis and identification of various compounds that can be obtained by using the mass spectrometer as the detector for a gas chromatograph suggest that this type of compound instrument would be of great use in many quality control and research laboratories. It extends the usefulness of both the gas chromatograph and the mass spectrometer.

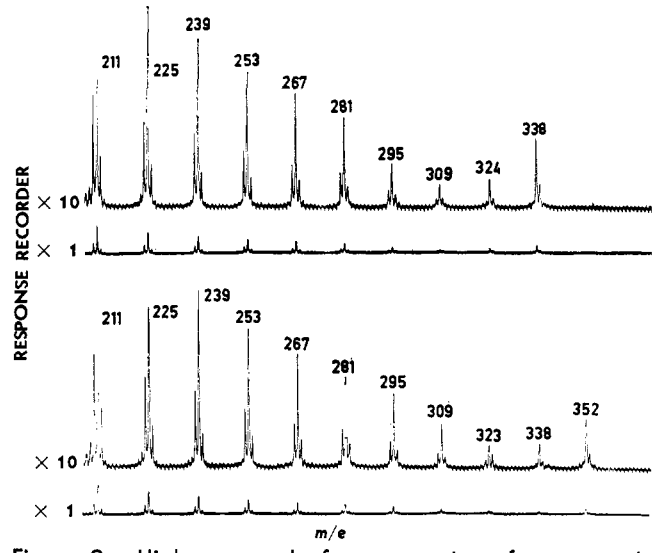

Figure 9. High-mass end of mass spectra of components C_{24} and C_{25} from chromatogram of paraffin (Figure 8)

The compounds were identified as *n*-tetracosane (mol. wt. 238) and *n*-pentacosane (mol. wt.) 352

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LITERATURE CITED

(1) Becker, E. W., "Separation of Isotopes," p. 360, George Newnes Ltd., London, 1961.
(2) Biemann, K., "Mass Spectrometry, Organic Chemical Applications," p. 172, McGraw-Hill, New York, 1962.
(3) Brunnée, C., Jenckel, L., Kronenberger, K., ASTM Committee E-14 Meeting, New Orleans, La., 1962.
(4) Dorsey, J. A., Hunt, R. H., O'Neal, M. J., ANAL. CHEM. **35**, 511 (1963).
(5) Ebert, A. A., *Ibid.*, **33**, 1865 (1961).
(6) Gerson, T., Hawke, I. C., Shortland, F. B., *Biochem. J.* **74**, 366 (1960).
(7) Gohlke, R. S., ANAL. CHEM. **31**, 535 (1959).
(8) Haathi, E., *Scand. J. Clin. Lab. Invest.* **13**, suppl. 59 (1961).
(9) Hansen, R. P., Shorland, F. B., Cooke, J. N., *Biochem. J.* **77**, 64 (1960).
(10) Horning, E. C., Moscatelli, E. A., Sweeley, C. C., *Chem. Ind.* **1959**, 751.
(11) Lindeman, L. P., Annis, J. L., ANAL. CHEM. **32**, 1742 (1960).
(12) Ryhage, R., *Arkiv. Kemi* **20**, 185 (1962).
(13) Ryhage, R., Ställberg-Stenhagen, S., Stenhagen, E., *Ibid.*, **14**, 247 (1959).
(14) Ryhage, R., Ställberg-Stenhagen, S., Stenhagen, E., Karolinska Institutet, Stockholm, Sweden, unpublished data, 1961.
(15) Ryhage, R., Stenhagen, E., *Arkiv Kemi* **13**, 523 (1959).
(16) *Ibid.*, **15**, 291 (1960).
(17) Ryhage, R., Sydow, E. von, *Acta Chem. Scand.* **17**, 2025 (1963.)
(18) Váradi, P., Ettre, K., ANAL. CHEM. **34**, 1417 (1962).
(19) Váradi, P., Ettre, K., *Ibid.*, **35**, 410 (1963).

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Reaction of Ethers with Acetyl Chloride and the Identification of Products by Gas Chromatography

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▶ **The reaction of simple and polymeric ethers with acetyl chloride in the presence of a strong Lewis acid salt has been studied. Reaction products have been isolated and identified as the corresponding chloroacetates. Optimum conditions for quantitative conversion have been found and utilized to develop a procedure for the classification of ethers. The procedure has been applied to polyglycol ethers containing, in some instances, mixed ether groups. Gas liquid chromatography is used for the determination of reaction products. Colored compounds are formed during the reaction which appear to be linearly related to the concentration of the polyether. Possibilities of a new colorimetric method are presented.**

POLYOXYALKYLENE polymers of various compositions are commercially available and find many applications. These materials usually are made by ethoxylation and/or propoxylation of low molecular weight alcohols. The degree of hydroxyl functionality of the alcohol controls the number of terminal hydroxyl groups in the resulting polyether. Consequently, mono-, di-, and trihydroxy polyethers are available. For some applications, the hydroxyl groups are reacted to form the polyether alkoxy compound, where usually the terminal alkyl group is C_1–C_9. Generically, the compounds can be represented by the following formula;

$$R_1\left[O(CH_2CH_2O)_x(CH_2\underset{\displaystyle CH_3}{\underset{|}{C}}HO)_yR_2\right]_z$$