

CHROM. 19 227

IDENTIFICATION OF ω -OXOCARBOXYLIC ACIDS AS ACETAL ESTERS IN AEROSOLS USING CAPILLARY GAS CHROMATOGRAPHY-MASS SPECTROMETRY

KIMITAKA KAWAMURA* and R. B. GAGOSIAN

Department of Chemistry, Woods Hole Oceanographic Institution, Woods Hole, MA 02543 (U.S.A.)

(First received September 12th, 1986; revised manuscript received November 6th, 1986)

SUMMARY

An homologous series of ω,ω -dimethoxycarboxylic acid methyl esters (acetal esters) has been identified in the most polar methyl ester fraction separated from remote aerosol samples by liquid chromatography. These compounds were separated by silica gel column chromatography and their structures determined by capillary gas chromatography (GC) and GC-mass spectrometry with a synthesized standard. The acetal esters are originally present in the samples as ω -oxocarboxylic acids, which are derivatized to acetal esters during treatment with boron trifluoride in methanol. This method enables the determination of ω -oxocarboxylic acids in environmental samples as their acetal esters.

INTRODUCTION

During a search for terrestrial biological source markers in remote marine aerosols, we found an homologous series of previously unreported organic compounds in aerosol, rain and surface seawater samples¹. These compounds appeared as major species in the most polar carboxylic acid methyl ester liquid chromatographic fraction, indicating that they were carboxylic acids containing additional functional groups. Electron-impact mass spectra of the homologous series showed an intense parent ion at m/z 75 with major fragments at m/z $159 + 14n$, where $n = 0-5$. The isolation and structural elucidation of these compounds and the determination of their sources and fate are important in furthering our understanding of the cycling of organic compounds in the atmosphere and ocean surface.

In this paper we describe the isolation and structural determination of these compounds in remote samples by using a capillary gas chromatography-mass spectrometry (GC-MS) method.

EXPERIMENTAL

Reagents and materials

All organic solvents were obtained from Burdick & Jackson. They were redis-

tilled in all-glass apparatus and checked for purity by GC prior to use. Water free from organic compounds was prepared by oxidation of distilled water with potassium permanganate, followed by redistillation. To minimize organic contaminants, potassium hydroxide pellets (Baker Chemical Co.) were washed with methanol under sonication (10 s, twice) and then dissolved in fresh methanol to give a 0.1 *M* solution. A 6 *M* hydrochloric acid solution was extracted with dichloromethane and hexane prior to use. Boron trifluoride in methanol (Supelco, 14%) was purified by extraction with hexane. Aleuritic acid (9,10,16-trihydroxypalmitic acid) and sodium periodate were obtained from Fluka, *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (MNNG) and a MNNG-diazomethane kit from Aldrich Chemical Co.

Silica gel (Bio-Sil A, 200–400 mesh; Bio-Rad Labs.) was extracted with methanol and dichloromethane, dried under vacuum using a rotary evaporator (water-bath temperature gradually increased from 25 to 80°C) and heated in an oven at 210°C for 24 h. The silica gel was deactivated with 1% water. After occasional shaking for 2 days, the gel was stored in hexane.

Samples

Aerosol samples were collected with Gelman Type A/E glass fiber or Pallflex type quartz fiber filters using Hi-vol samplers², from the top of a 20-m tower built on Enewetak Atoll, Marshall Island³, from the top of a 8-m tower aboard the R/V Moana Wave on a cruise in the central North Pacific or from 1 m above ground level on American Samoa. Blank filters were exposed to the air in the sample shelters during sample collection. Details of the sample collection and handling are described elsewhere^{2,4}.

Procedures

One half of the filter was extracted with 100 ml of 0.1 *M* methanolic potassium hydroxide under reflux for 2 h. The alkaline solution was then poured off and the remaining filter extracted further with methanol and dichloromethane. The extracts were combined and concentrated to *ca.* 2 ml by rotary evaporation under vacuum. A 10-ml volume of redistilled water was then added. Neutral compounds were first extracted with hexane–dichloromethane (2:1) (8 ml × 3), then the remaining alkaline solution was acidified to pH ≈ 1 with 6 *M* hydrochloric acid. Carboxylic acids were extracted with dichloromethane (8 ml × 3) and the dichloromethane layer was back-washed with redistilled water. The acids were methylated with 0.5 ml of 14% methanolic boron trifluoride in a bath of boiling water for 30 min. Hexane (8 ml) was then added to extract the methyl esters. The hexane extract was washed with distilled water (5 ml × 3) and separated into three fractions on a silica gel column (Pasteur pipette, height 4 cm, *ca.* 1 ml of gel in hexane) by eluting with (1) 5 ml hexane–dichloromethane (1:2) (monocarboxylic acid methyl esters), (2) 9 ml dichloromethane–ethyl acetate (98:2) (dicarboxylic acid dimethyl esters) and (3) 4 ml dichloromethane–methanol (95:5) (acetal and hydroxyacid methyl esters). The third fraction containing the acetal esters was concentrated under nitrogen and analyzed by GC.

GC and GC-MS

A Carlo Erba 4160 gas chromatograph equipped with an on-column injector and a flame ionization detector was used with a fused-silica DB-5 capillary column

(30 m \times 0.32 mm, J&W Scientific, Folsom, CA, U.S.A.). Hydrogen was used as a carrier gas. The column temperature was programmed from 80 (2 min) to 310°C (10 min) at 5°C/min; data were processed with a Varian Vista 402. GC-MS analyses were conducted with a Carlo Erba 4160 gas chromatograph interfaced to a Finnigan 4500 mass spectrometer. The data were processed with an INCOS 2300 data system. The GC conditions were similar to those mentioned above, except that helium was used as a carrier gas.

Synthesis

ω -Oxononanoic acid was synthesized by oxidizing aleuritic acid with sodium periodate⁵. Aleuritic acid (151 mg) was dissolved in 1 ml of 0.5 M sodium hydroxide solution and 1 ml chloroform under sonication and treated with sodium periodate (134 mg) at 20°C for 30 min. The reaction mixture was filtered and diluted in 10 ml water. The solution was first extracted with hexane-dichloromethane (75:25) (10 ml \times 3) and then acidified to pH \approx 1 with 6 M hydrochloric acid. The acidic solution containing ω -oxononanoic acid was extracted with hexane-dichloromethane (75:25) (10 ml \times 2), washed with water and concentrated. Formation of ω -oxononanoic acid was confirmed by GC-MS after derivatization of the carboxyl group with diazomethane. The mass spectra of methyl ω -oxononanoate showed fragment ions of m/z 41, 43, 55, 59, 69, 74, 83, 87, 111, 143, 155, 158, 168 and 185, in agreement with literature values⁶. Fragment ions of m/z 74, 87 and 143 are characteristic of methyl esters and those of m/z 158 (M - CO), 168 (M - H₂O) and 185 (M - 1) are ascribed to the presence of a formyl group⁷. Methyl ω -oxononanoate was then treated with methanolic boron trifluoride (100°C, 30 min) to yield methyl 9,9-dimethoxynonanoate by converting the aldehyde group into an acetal.

RESULTS AND DISCUSSION

Fig. 1a shows a typical gas chromatogram of the unknown compounds isolated from the aerosol samples. Major peaks appeared early in the chromatogram. Whole procedure blanks (including sample collection, transportation, storage and analytical procedures) showed that the peaks are not due to contamination. Although this fraction should contain hydroxyacid methyl esters, the major components are not α -, β - or ω -hydroxyacid esters. From their chromatographic behavior on silica gel, they should be carboxylic acid methyl esters containing an additional functional group.

Fig. 2 presents the electron-impact mass spectra of the three dominant compounds in Fig. 1a. A strong mass fragment of m/z 75 was observed for these major and other species, suggesting that they are compounds of an homologous series. The mass fragment of m/z 75 has been reported for (CH₃O)₂CH⁺ which originates from 1,1-dimethoxypropane⁸. Therefore, the compounds of the homologous series were hypothesized to be carboxylic acid methyl esters containing a terminal dimethoxy (acetal) functional group. The dominant species (compound C in Figs. 1 and 2) was proposed to be methyl 9,9-dimethoxynonanoate (MW 232), since a fragment of m/z 201 is equivalent to M - 31: a loss of CH₃O from either a methyl ester or an acetal and the fragment of m/z 231 (M - 1) suggests a loss of the hydrogen atom on the 9-carbon (acetal portion)⁶. As expected, the molecular ion was not observed^{8,9}. By

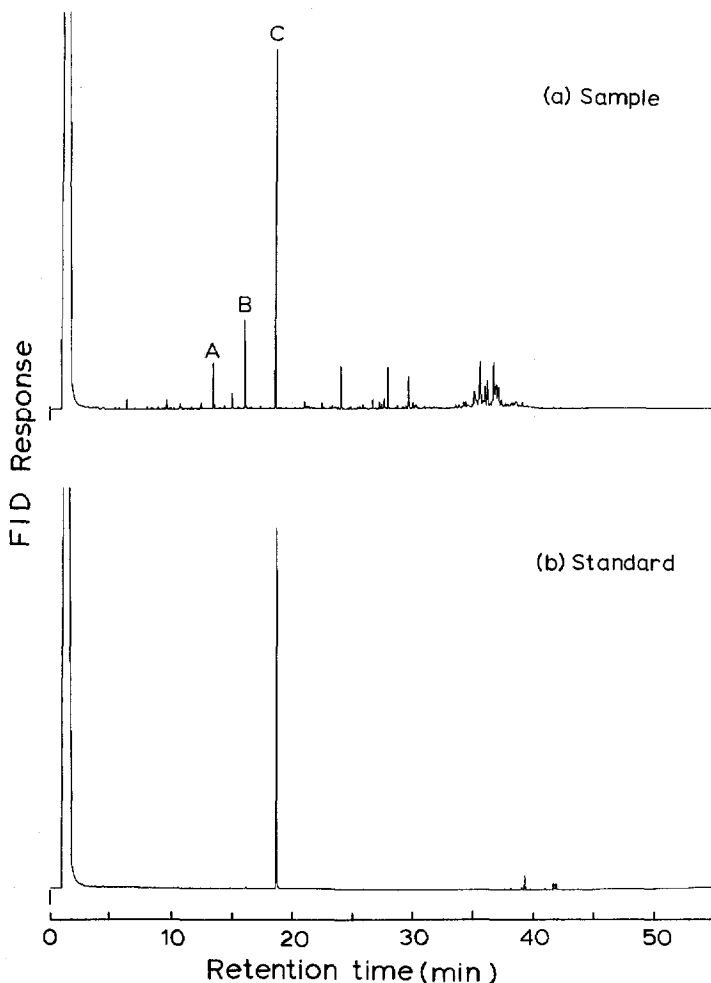


Fig. 1. Reconstructed capillary gas chromatograms of (a) unknown compounds from a Samoa aerosol sample (SMAS 3) and (b) an authentic standard of methyl 9,9-dimethoxynonanoate.

analogy, compounds A and B (see Figs. 1a and 2) were proposed to be methyl esters of 7,7-dimethoxyheptanoic acid and 8,8-dimethoxyoctanoic acid, respectively.

For a final structural identification, methyl 9,9-dimethoxynonanoate was synthesized as described in the Experimental section. Fig. 1b shows the gas chromatogram of synthesized methyl 9,9-dimethoxynonanoate. Comparison of the retention time of the authentic compound to that of the sample (see Fig. 1a, b), coinjection of the authentic standard with the sample (SMAS 3) and matching of the mass spectrum of the unknown with that of the synthesized compound (see Figs. 2c and 3) confirmed the identification of the major peak as methyl 9,9-dimethoxynonanoate.

We suggest that the acetal esters identified were originally present in the aerosol samples as ω -oxocarboxylic acids. They were observed as acetal methyl esters because

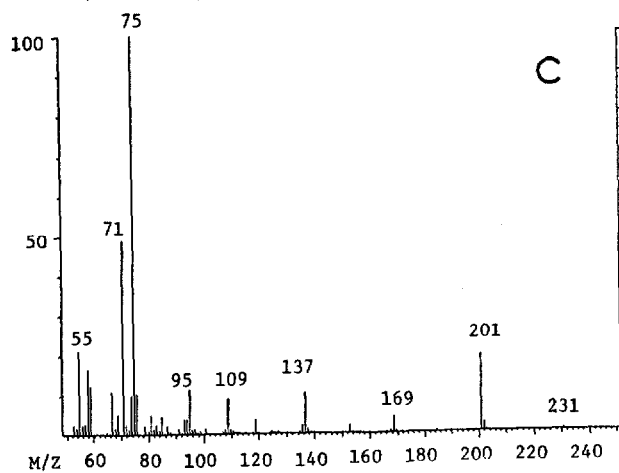
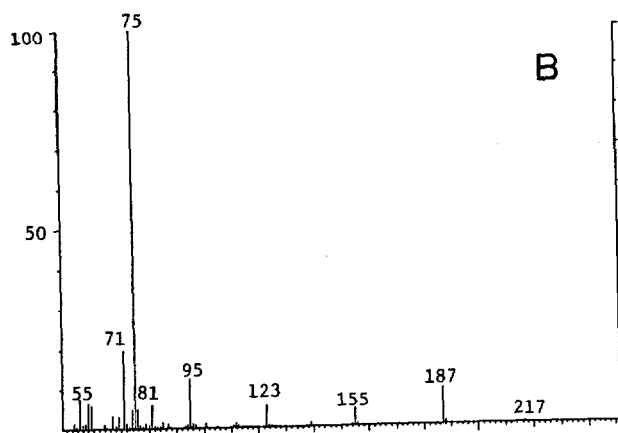
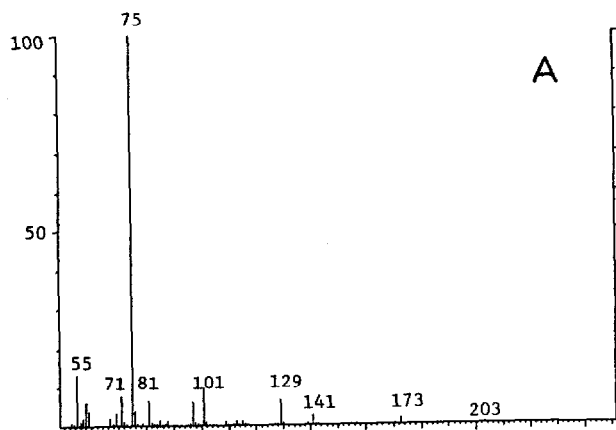
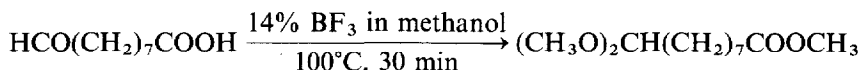


Fig. 2. Mass spectra of unknown compounds in peaks A, B and C (see Fig. 1a).

during work-up, the methanolic boron trifluoride quantitatively catalyzed the following reaction:



In order to prove that the underivatized ω -oxocarboxylic acids were present in the samples, one-eighth of the aerosol (SMAS 3) filter was analyzed by the procedure described above, except that diazomethane in diethyl ether (containing 6% methanol) was used as a methylating agent, instead of the boron trifluoride. Diazomethane methylates carboxylic acids but not aldehydes. GC and GC-MS analysis showed the ω -oxocarboxylic acid methyl esters (C_9 being major) in the second and third fractions eluted from the silica gel column using the same solvent system as mentioned above. A small amount (*ca.* 10%) of an acetal ester (corresponding to ω -oxononanoic acid) was also detected due to free hydrogen derived from organic acids in the acidic fraction¹⁰. Other steps in the analytical procedure were also checked to make sure that the corresponding acetal ester was not formed during its isolation and purification; none was found. These results indicate that ω -oxocarboxylic acids are present in our samples and are converted into their corresponding acetal esters by the analytical scheme used.

Based on their characteristic mass fragments (m/z 75, $M - 31$, $M - 1$), C_6 - C_{11} ω -oxocarboxylic acids were identified as acetal esters in the aerosol samples. These compounds have not previously been reported in environmental samples. To our knowledge, ω -oxononanoic acid (which is the dominant species in the samples) has not been reported in plants or other organisms. They are probably not of an-

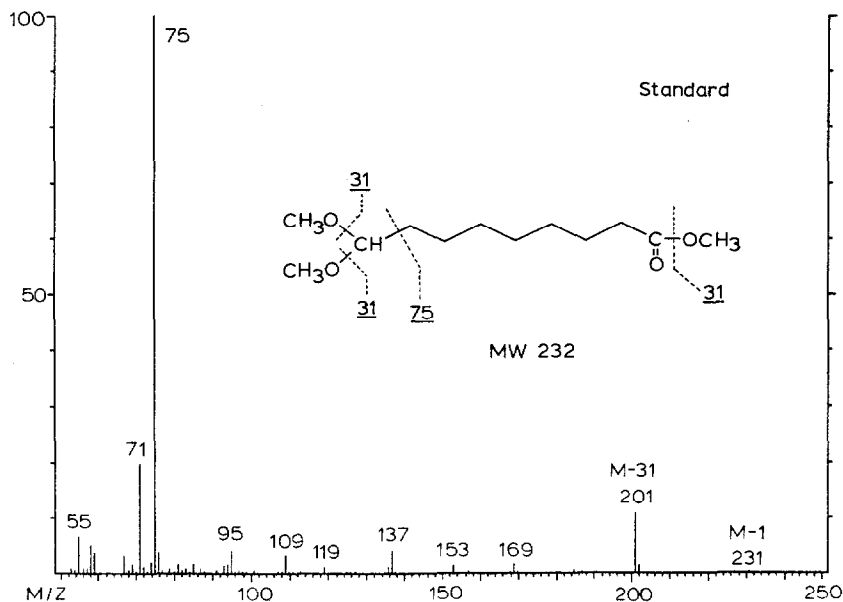


Fig. 3. Mass spectrum of synthesized methyl 9,9-dimethoxynonanoate.

thropogenic origin because other compound classes isolated from the samples showed no anthropogenic input^{3,4}. Rather, we believe that they are secondary reaction products from certain biogenic compounds such as unsaturated fatty acids in the atmosphere and surface seawater¹.

CONCLUSIONS

ω -Oxocarboxylic acids can be isolated as ω,ω -dimethoxycarboxylic acid methyl esters (acetal esters) from environmental samples and their structures determined by capillary gas chromatography-mass spectrometry. The acetal esters were derived from ω -oxocarboxylic acids during treatment with methanolic boron trifluoride and were separated by silica gel column chromatography. Since the mass spectra of acetal esters of ω -oxo acids give a characteristic base peak at m/z 75, mass chromatography at m/z 75 is a potentially important technique in detecting ω -oxocarboxylic acids in environmental samples.

ACKNOWLEDGEMENTS

We thank E. T. Peltzer, F. A. Mendoza, S. C. Brassell and J. K. Volkman for their useful discussions. The aerosol samples were collected by E. T. Pelzer and J. B. Alford. Mass spectral data acquisition was performed by N. M. Frew and C. G. Johnson. This study was conducted as a part of the SEAREX program supported by NSF Grant OCE 84-06666. Woods Hole Oceanographic Institution Contribution No. 6286.

REFERENCES

- 1 K. Kawamura and R. B. Gagosian, *Nature (London)*, in press.
- 2 E. T. Peltzer, J. B. Alford and R. B. Gagosian, *Woods Hole Oceanographic Institution Technical Report*, WHOI-84-9 (1984).
- 3 R. B. Gagosian, O. C. Zafiriou, E. T. Peltzer and J. B. Alford, *J. Geophys. Res.*, 87 (1982) 133.
- 4 R. B. Gagosian, in P. Buat-Menard (Editor), *The Role of Air/Sea Exchange in Geochemical Cycling*, Reidel, Dordrecht, 1986, pp. 409-442.
- 5 I. Ujvary, A. Kis-Tamas and L. Novak, *J. Chem. Ecol.*, 11 (1985) 113.
- 6 H. Budzikiewicz, C. Djerassi and D. H. Williams, *Mass Spectrometry of Organic Compounds*, Holden-Day, San Francisco, CA, 1967, p. 690.
- 7 S. D. Killops, *Water Res.*, 20 (1986) 167.
- 8 R. A. Friedel and A. G. Sharkey, Jr., *Anal. Chem.*, 28 (1956) 940.
- 9 W. H. McFadden, J. Wasserman, J. Corse, R. E. Lundin and R. Teranishi, *Anal. Chem.*, 36 (1964) 1030.
- 10 J. March, *Advanced Organic Chemistry*, Wiley, New York, 1984, p. 1346.