1455, 1260, 1027 cm^{-1}; exact mass, calcd for $C_9H_{10}O_2$ 150.06808, found 150.06918.

Phenol **6** (mp 118–120 °C) was purified from the high pH solvolysis mixture by column chromatography (silica gel, EtOAc). Data for **6**: ^1H NMR ($CDCl_3/CD_3SOCD_3$) δ (Me_4Si) 2.6–3.4 (m, 4 H), 4.60 (m, 2 H), 6.4–7.2 (m, 3 H), 8.33 (s, 1 H); IR (KBr) 3400 (br), 2950, 1590, 1472, 1285, 1050 cm^{-1}; exact mass calcd for $C_9H_{10}O_2$ 150.06808, found 150.06939.

The structures of phenols **6** and **7** follow unambiguously by comparison of their ^1H NMR spectra with the published[9] spectra of 4-indanol and 5-indanol, respectively.

Addition of LiSMe to Arene Oxide 1. Synthesis and Characterization of Thioethers 10 and 11. To arene oxide **1** (0.456 g, 3.04 mmol) in *tert*-butyl alcohol (5 mL) was added LiSMe (0.56 g, 10.37 mmol) in H_2O (5 mL) and the mixture was stirred 45 min at ambient temperature. Extraction (4 × 25 mL of Et_2O), drying ($MgSO_4$) and removal of solvent gave a mixture (0.58 g, 97%) containing dihydro aromatics **8** and **9**. The NMR spectrum of the mixture is complex but shows (270 MHz, $CD_3SOCD_3/CDCl_3$) olefinic absorptions [δ (Me_4Si) 5.6–6.2] and two clearly discernible singlets attributable to –SMe bound to saturated carbon at δ (Me_4Si) 1.88 and 1.83 in the ratio of 2.5:1, respectively. Upon acidification (CF_3COOH) or upon standing overnight the mixture aromatized to **10** + **11**. The ^1H NMR spectrum (270 MHz, $CD_3SOCD_3/CDCl_3$) of **10** + **11** lacks absorptions in the region δ (Me_4Si) 5.6–6.2 and shows one predominant –SMe peak (singlet) at δ (Me_4Si) 2.38.

Column chromatographic separation of **10** and **11** was achieved on silica gel (35 g) using 3:1 hexanes/EtOAc as eluent. The first eluting compound (110 mg, 20%) was shown (vide infra) to be thioether **10**, mp 56–57 °C. Data for **10**: ^1H NMR ($CDCl_3$) δ (Me_4Si) 2.38 (s, 3 H), 2.93 (m, 5 H), 4.55 (m, 1 H), 6.96 (m, 3 H); IR (KBr) 3300 (br), 2942, 1580, 1455, 1442, 1028 cm^{-1}; exact mass calcd for $C_{10}H_{12}OS$ 180.06089; found 180.06147. Anal. Calcd: C, 66.63; H, 6.71. Found: C, 66.63; H, 6.90. A middle eluting fraction (40 mg) was shown (TLC) to be a mixture of **10** + **11**. The last eluting fraction (124 mg, 23%) was shown (vide infra) to be thioether **11**, mp 67–69 °C. Data for **11**: ^1H NMR ($CDCl_3$) δ (Me_4Si) 2.38 (s, 3 H), 2.93 (m, 5 H), 4.55 (m, 1 H), 6.95 (br s, 3 H); IR (KBr) 3295 (br), 2950, 1595, 1568, 1480, 1422, 1036 cm^{-1}; exact mass calcd for $C_{10}H_{12}OS$ 180.06089, found 180.06305. The combined yield of material eluted from the column was 50%; the remaining material was not accounted for.

Synthesis of Sulfonium Salts 12 and 13. Structure Assignments for Thioethers 10 and 11. Separated thioethers **10** and **11** were converted to **12** and **13**, respectively, by the procedure given below for **10** → **12**.

To thioether alcohol **10** (160 mg, 0.889 mmol) in dimethylformamide (0.7 mL) was added NaH (ca. 40 mg, 1.7 mmol). When evolution of H_2 ceased, MeI (55 μL, 0.89 mmol) was added and the mixture was stirred overnight. Removal of solvent, extractive workup, and preparative TLC gave the methyl ether derived from **10** (158 mg, 92%).

The methyl ether (147 mg, 0.757 mmol) was converted to sulfonium salt **12** by treatment with $Me_3O^+ BF_4^-$ (117 mg, 0.793 mmol) in CH_2Cl_2 (0.9 mL) over a period of 30 h. Removal of solvent and trituration with Et_2O yielded sulfonium salt **12** (167 mg, 75%) as light purple crystals (mp 147–148 °C).

Similar steps produced sulfonium salt **13** (mp 82–83 °C) from thioether alcohol **11**.

Acknowledgment is made to the National Institutes of Health for support of this work. We also thank Dr. C. E. Costello for mass spectra.

Registry No. **1**, 70897-83-5; **2**, 7567-68-2; *anti*-**3**, 70897-84-6; *syn*-**3**, 70954-00-6; **4**, 70897-85-7; **5**, 70897-86-8; **6**, 70897-87-9; **7**, 51927-77-6; **8**, 70897-88-0; **9**, 70897-89-1; **10**, 70897-90-4; **11**, 70897-91-5; **12**, 70897-93-7; **13**, 70897-95-9.

(9) "The Aldrich Library of NMR Spectra", Vol. IV, C. J. Pouchert and J. R. Campbell, Ed., Aldrich Chemical Co., Milwaukee, Wisconsin 1974, spectra numbers 120-D and 121-A.

Preparation and Purification of Arachidonic Acid Hydroperoxides of Biological Importance

Ned A. Porter,* James Logan, and Voula Kontoyiannidou

Paul M. Gross Chemical Laboratories, Duke University, Durham, North Carolina 27706

Received March 22, 1979

Singlet oxygen oxidation of arachidonic acid (5,8,11,14-eicosatetraenoic acid) leads to eight hydroperoxides that may be separated by high-pressure liquid chromatography. The hydroperoxides result from allylic oxidation of one of the double bonds of the polyene fatty acid, a trans double bond being formed in the process. 12-(Hydroperoxy)eicosatetraenoic acid, 12-HPETE, a biologically important hydroperoxide formed from arachidonic acid and a lipoxygenase enzyme present in blood platelets, may be prepared by this approach.

Arachidonic acid (5,8,11,14-eicosatetraenoic acid, 20:4) reacts with molecular oxygen in reactions catalyzed by two distinctly different enzymes found in platelets (Figure 1). The first enzyme, cyclooxygenase, converts arachidonic acid into the prostaglandin (PG) endoperoxides, PGG_2 and PGH_2.[1,2] The PG endoperoxides are enzymatically converted to the thromboxanes[3] and other nonperoxidic PG's,[4] and these prostaglandins and thromboxanes play an important role in the chemistry associated with the aggregation of platelets.

A second enzymatic pathway of arachidonic acid in platelets was revealed when it was shown that a platelet lipoxygenase converts arachidonic acid into 12-(hydroperoxy)eicosatetraenoic acid, 12-HPETE.[5,6] Although the biological function of 12-HPETE has not been completely defined, lipid hydroperoxides such as 12-HPETE have been recently shown to be important mediators of diverse

(1) Hamberg, M. Samuelsson, B. *Proc. Natl. Acad. Sci. U.S.A.* **1973**, *70*, 899.
(2) Nugteren, D. H.; Hazelhof, E. *Biochim. Biophys. Acta* **1973**, *326*, 448.
(3) Hamberg, M.; Svensson, J.; Samuelsson, B. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 2994.
(4) Nicolaou, K. C.; Burnette, W. E. *Angew. Chem., Int. Ed. Engl.* **1978**, *17*, 293.
(5) Nugteren, D. H. *Biochim. Biophys. Acta* **1975**, *380*, 299.
(6) Hamberg, M.; Samuelsson, B. *Proc. Natl. Acad. Sci. U.S.A.* **1974**, *71*, 3400.

0022-3263/79/1944-3177$01.00/0

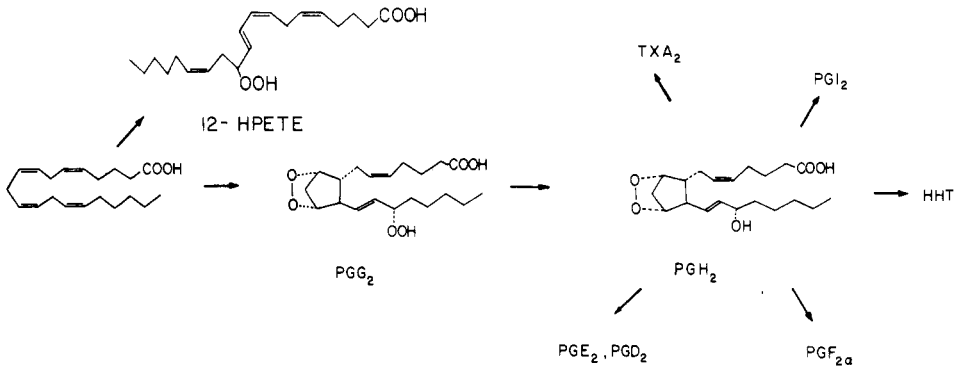


Figure 1. Arachidonic acid oxidation by platelets.

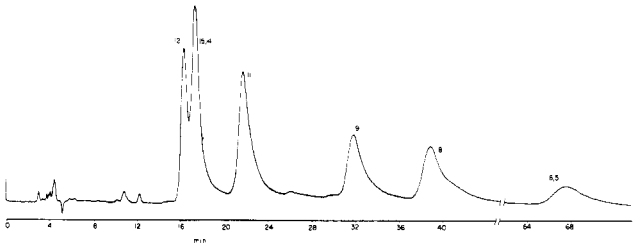


Figure 2. High-pressure LC trace of HPETE isomers.

biological processes. For example, lipid hydroperoxides have been shown to (1) inhibit the formation of PGI_2 from PGH_2,[7] (2) enhance anaphylactic mediator release from guinea pig lungs,[8,9] (3) activate guanylate cyclase,[10,11] and (4) inhibit the aggregation of human platelets.[12]

Because of the biological importance of lipid hydroperoxides, we sought to develop synthetic approaches to this class of compound. To date, the only method available for the preparation of lipid hydroperoxides is via the enzymatic route,[5,13] and this approach leaves little flexibility in product structure. The platelet enzyme gives only the 12-hydroperoxy fatty acid,[16] and the lipoxygenase from soybeans gives only the 15-hydroperoxy compound.[14,15] We report here on the preparation of arachidonic acid hydroperoxides by singlet oxygen oxidation of the fatty acid. This method gives a mixture of all the possible allylic hydroperoxides, and these hydroperoxides may be purified by preparative high-pressure liquid chromatography (LC).

Results and Discussion

A methanol solution of arachidonic acid and methylene blue was photolyzed under an oxygen atmosphere. Progress of the photolysis was followed by thin-layer chromatography, and new peroxidic products were formed as arachidonic acid was consumed. A typical thin-layer chromatogram of the photolysis mixture showed four peroxidic fractions (TLC A–D) that contained lipid hydroperoxides (TLC A, R_f 0.21; TLC B, R_f 0.18; TLC C, R_f 0.15; TLC D, R_f 0.07). Column chromatography of the reaction product mixture at 0 °C on silica gel separated the polar peroxide of TLC D from the hydroperoxide in TLC A–C, the hydroperoxide compounds being isolated in 35–40% yield under conditions of optimal conversion.

Table I. Elution Order of the Isomeric HPETE, HETE, HPETE Methyl Esters, and HETE Methyl Esters on 10 μm Silica Gel

derivative	elution order[a]
HPETE	12, (15, 14), 11, 9, 8, (5, 6)
HPETE methyl ester	15, (14, 12), 11, (9, 8), (5, 6)
HETE	12, 15, 11, 9, 8, (5, 6)
HETE methyl ester	12, (15, 14), 11, 9, 8, (5, 6)

[a] Parentheses indicate incomplete separation of isomers.

LC of the combined TLC A–D compounds on a semipreparative 10-μm silica gel column indicated that six fractions (fractions I–VI) detectable by a UV detector operating at 235 nm were present in the product mixture (Figure 2). TLC fraction A was shown to contain the first three fractions eluting from the LC column, TLC B only the fourth LC fraction, TLC C only the fifth LC fraction, and TLC D only the sixth LC fraction.

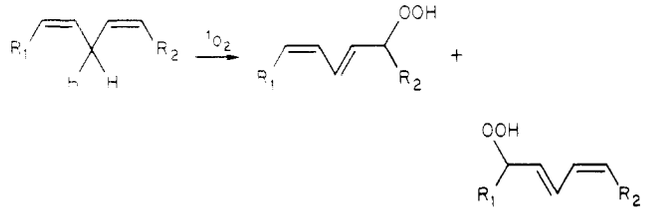
Consideration of the possible allylic hydroperoxides formed from arachidonic acid suggests that six of the eight possible hydroperoxide positional isomers (at carbons 5, 8, 9, 11, 12, and 15) would have a conjugated diene structure whereas two of the product hydroperoxides (at carbon 6 and 14) would be nonconjugated polyolefins. The position of the hydroperoxide functionality on the fatty acid chain of the product hydroperoxide was determined as follows. The hydroperoxy fatty acids were esterified with diazomethane and hydrogenated over platinum, affording the saturated hydroxy fatty acid methyl esters. The alcohol functional group was trimethylsilylated and this silylated methyl ester was then analyzed by gas chromatography–mass spectrometry. The characteristic fragmentation of the carbon chain at the bond adjacent to the trimethylsiloxy group indicates the position of oxygen substitution in the original HPETE isomer.[16] By the use of this method of analysis, LC fractions I–VI (Figure 2) were shown to contain fatty acid hydroperoxides substituted as follows: I, 12; II, 15 and 14; III, 11; IV, 9; V, 8; VI, 5 and 6.

UV spectroscopy of the purified LC fractions indicated absorption λ_{max} at 235 nm with molar absorbtivity values of 23 000–30 000 for all of the hydroperoxides. This 235-nm absorption is characteristic of conjugated dienes, R—CH=CH—CH=CH—R′.[17] Infrared spectroscopy of the

(7) Gryglewski, R.; Bunting; Moncada, S.; Flower, R.; Vane, J. *Prostaglandins* **1976**, 12, 685.
(8) Adcock, J. J.; Garland, L. G.; Moncada, S.; Salmon, J. A. *Prostaglandins* **1978**, *16*, 163.
(9) Adcock, J. J.; Garland, L. G.; Moncada, S.; Salmon, J. A. *Prostaglandins* **1978**, 16, 179.
(10) Hidaka, H.; Asano, T. *Proc. Natl. Acad. Sci. U.S.A.* **1977**, *74*, 3657.
(11) Graff, G.; Stephenson, J. H.; Glass, D. B.; Maddox, M. K.; Goldberg, N. D. *J. Biol. Chem.* **1978**, *253*, 7662.
(12) Bild, G. S.; Bhat, S. G.; Axelrod, B.; Iatridis, P. G. *Prostaglandins* **1978**, *16*, 795.
(13) Hamberg, M.; Samuelsson, B. *J. Biol. Chem.* **1967**, *242*, 5329.
(14) Hamberg, M.; Samuelsson, B.; Björkhem, I.; Danielsson, H. "Molecular Mechanisms of Oxygen Activation"; Hayaishi, I., Ed.; Academic Press: New York and London, 1974; Chapter 2.
(15) Funk, M. O.; Isaac, R.; Porter, N. A. *Lipids* **1976**, *11*, 113.
(16) Richter, W. J.; Burlingame, A. L. *Chem. Commun.* **1968**, 1158.
(17) Chan, H. W. S.; Levett, G. *Lipids* **1977**, *12*, 99.

column-chromatographed mixture of hydroperoxides or of LC fraction III (11-OOH) showed absorptions at 3400, 2940, 1740, 985, and 950 cm^{-1}. The absorptions at 950 and 985 cm^{-1} indicate the presence of a trans,cis-substituted conjugated diene.[18]

Authentic 15-(hydroperoxy)eicosatetraenoic acid (15-HPETE)[15] and 12-HPETE[19] were compared to the synthetic hydroperoxides prepared by the singlet oxygen method. 15-HPETE prepared from arachidonic acid oxygenation catalyzed by soybean lipoxygenase[14] co-chromatographed with LC fraction II and TLC A. 12-HPETE prepared from platelet lipoxygenase[19] was chromatographically identical with LC fraction I and TLC A. It should be noted that enzymatically prepared 15-HPETE and 12-HPETE both have been assigned trans,cis stereochemistry at the conjugated diene unit. We thus conclude that the singlet oxygen oxidation of *cis*-polyene fatty acids yields hydroperoxide products that contain the trans,cis-conjugated diene functional group. Not only does infrared spectroscopy indicate the presence of trans,cis-conjugated double bonds but also the products of singlet oxygen oxidation compare exactly (high pressure LC, GC–MS, UV) with biological samples that are known to have the trans,cis diene.

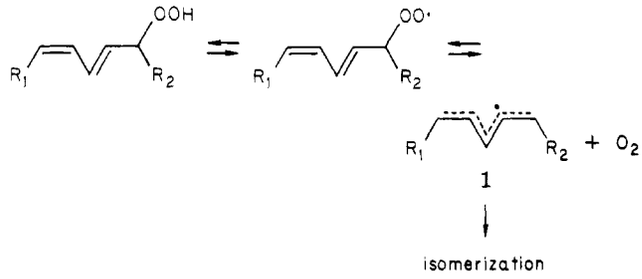


It should be noted that of the eight possible hydroperoxides formed from arachidonic acid two of these, the 14 and 6 isomers, have no conjugated diene and are not detectable by UV analysis. By chance, these isomers co-chromatograph with the UV-active 15- and 5-substituted compounds.

The net reaction observed in the singlet oxygen reaction is the conversion of one of the cis olefin units of the polyene into a trans-substituted allylic hydroperoxide. Simple monoolefins are known to form trans allylic hydroperoxides exclusively when reacted with singlet oxygen, and the products observed with the polyene fatty acids are thus not entirely unanticipated.[20,21]

The hydroperoxides appear to be reasonably stable if kept at 0 °C or below in concentrated hexane or ether solutions. If dilute solutions are kept at room temperature, considerable decomposition of the hydroperoxides occurs. Further, the initial column chromatography of the crude oxidation product mixture must, of necessity, be carried out at 0 °C or below. If this precaution is not taken, significant material loss of the primary HPETE isomers is noted. If care is not taken to keep the hydroperoxide isomers cold, isomerization of the trans,cis-conjugated diene to the more stable trans,trans structure may occur.[22] The mechanism of this isomerization has recently been shown to involve β scission of the peroxy radical leading to molecular oxygen and the pentadienyl radical **1**.[23]

Following equilibration of 1 with other stereochemical forms, re-formation of hydroperoxide would yield mixtures of geometric isomers, the trans,trans compound being thermodynamically preferred.



The hydroperoxy fatty acids (HPETE) were converted to the methyl esters (HPETE methyl ester) by reaction with diazomethane. Further, the hydroxy fatty acid derivatives could be prepared as the free acid (HETE) or methyl ester (HETE methyl ester) by triphenylphosphine reduction of the hydroperoxide group. The LC retention characteristics of these derivatives were examined, and the elution order of each of these fatty acid derivatives is presented in Table I.

The relative retention characteristics of the 12-substituted fatty acid derivatives is of interest. For unsaturated hydroperoxy fatty acids (HPETE) and the alcohols HETE as well as the HETE methyl esters studied here, maximum elution is observed with the C-12 isomer. Although the reasons for the relatively rapid migration of 12-HPETE, 12-HETE, and 12-HETE methyl ester on silica are not clear, one could speculate that specific hydrogen-bonding interactions between the hydroperoxy or alcohol and carboxylic acid or ester functional groups might enhance the migratory aptitude of the 12 isomer. This explanation seems unlikely, however, since with the HPETE methyl esters, the 12 isomer elutes after the 15 and 14 isomers. The relative retention on silica of C-20 fatty acid methyl esters containing alcohol or ketone substitution has been studied.[24] For the saturated C-20 alcohols and ketones, maximum elution is observed with alcohol or ketone substitution at C-14. The maximum elution of the C-12 isomer thus appears to be unique with the unsaturated hydroperoxy fatty acid and hydroxy fatty acid derivatives studied here.

It should be noted that singlet oxygen oxidation of 8,11,14-eicosatrienoic acid (20:3) also provides a mixture of unsaturated hydroperoxides. The elution order of the hydroperoxides derived from 20:3 is 12, (15, 14), (11, 9), 8. Here again, the 12-hydroperoxide is unusual in its relative order of elution, and conversion to the hydroperoxy methyl ester inverts the order of elution of the 15 and 12 isomers, a result that parallels our observations with the arachidonic acid hydroperoxides.

Although the method reported here does provide a straightforward route to the 12-, 11-, 9-, 8-, and 5-HPETE compounds of biological importance, the method is limited in that only milligram quantities of these compounds may be conveniently prepared by this method. The 12 isomer is particularly difficult to obtain in quantities greater than 1–2 mg because of the difficult separation between this isomer and the 15 and 14 isomers (Figure 1). With the semipreparative micrometer silica columns used in this study, acceptable separations were retained only with

(18) Chipanet, J. R.; Hawkins, J. M. *J. Am. Oil Chem. Soc.* **1959**, *36*, 535.
(19) We thank Dr. Marvin Siegel of Burroughs Wellcome Co. for a comparison of our product with authentic 12-HPETE.
(20) Orfanopoulas, M.; Grdina, M. B.; Stephenson, L. M. *J. Am. Chem. Soc.* **1979**, *101*, 275.
(21) Foote, C. S.; Denny, R. W. *J. Am. Chem. Soc.* **1971**, *93*, 5168.
(22) Singleton, J. A.; Pattee, H. E.; Nelson, M. S. *J. Am. Oil Chem. Soc.* **1978**, *55*, 377.
(23) Chan, H. W. S.; Levett, G.; Matthews, J. A. *J. Chem. Soc., Chem. Commun.* **1978**, 756.
(24) Morris, L. J.; Wharry, D. M.; Hammond, E. W. *J. Chromatogr.* **1968**, *33*, 471.
(25) Fieser, L. F.; Fieser, M. "Reagents for Organic Synthesis", Vol. 1; Wiley: New York, London, Sydney, 1967; p 191.

Table II. Retention Times for Eicosatetraenoic Acid Derivatives on High-Pressure LC

	position of substituent								flow rate, mL/	solvent		
corapd	15	14	12	11	9	8	6	5	min	acetic acid	2-propanol	hexane
HPETE	17[a]	17	16	22	32	39	68	68	4	1	8	991
HPETE methyl esters	14	15	15	16	20	20	26	26	3	1	6	993
HETE	9	9	8	14	25	31	70	70	5	1	10	989
HETE methyl esters	15	15	14	17	22	23	40	40	3	1	6	993

[a] Retention time in minutes.

Table III. Principal Fragments from Methyl(Trimethylsiloxy)eicosanoates

position of siloxy in C-20 chain	m/e for cleavage toward carbomethoxy	away from carbomethoxy
15	343	173
14	324	187
12	301	215
11	287	229
9	259	257
8	245	271
6	217	299
5	203	313

column loading of less than 5–7 mg total of HPETE compounds. On the other hand, the preparation of HPETE isomers reported here is only a one-step synthesis from a readily available starting material, and it may be possible to use newer preparative chromatography techniques in order to avoid the column-loading problem.

Experimental Section

Chromatography. A. Thin-Layer Chromatography. Precoated TLC plates (Merck, St. Louis, MO; silica gel 60F-254; 5 cm × 20 cm × 0.25 mm) were used without activation. Products were visualized by fluorescence quenching (conjugated diene), $Ce(SO_4)_2$ charring, and ferrous thiocyanate (hydroperoxides). The solvent used for elution of the arachidonic acid derivatives was acetic acid–2-propanol–hexane (1:20:229, v/v).

B. High-Pressure Liquid Chromatography. A Waters Associates 10 μm Porasil column (7.8 mm i.d. × 30 cm) was used for separation of the fatty acid derivatives. Solvents used for LC separations were acetic acid–2-propanol–hexane. The amount of 2-propanol varied from 0.4 to 1.2% depending on the polarity of the materials to be separated. The exact solvent conditions required for LC of each derivative are given in Table II. Both hexane and 2-propanol were distilled immediately before use.

Singlet Oxygen Oxidation of Arachidonic Acid. Arachidonic acid (99+%, Nu-Chek-Prep, Elysian, MN) was used without further purification. Methanol solvent was distilled before use. A 500-mL solution of 500 μL (450 mg, 1.48 mmol) of arachidonic acid and 180 mg of methylene blue in methanol was placed in a Pyrex photolysis well equipped with internal glass tubing (for cooling the vessel) and a side arm that allowed for bubbling of oxygen gas through the solution during photolysis. The reaction mixture was kept at −10 to 0 °C by the circulation of a cold ethylene glycol–water mixture through the spiral glass tubing.

Photolysis of the methanol solution with a 450-W medium-pressure mercury lamp (uranium-glass filter) was conducted for 24 h, during which time the progress of the photolysis was monitored by thin-layer chromatography. Four UV and $Fe(SCN)_2$ positive (peroxide) spots were observed at R_f 0.21, 0.18, 0.15, and 0.07 on the thin-layer analysis of the photolysate.

After photolysis was completed, the methanol was removed at 0.1 mmHg by flash distillation into a series of three traps, the first being a dry ice/acetone slurry and the last two being liquid nitrogen. The residue was taken up in 2 mL of 10% ethyl ether in hexane and was loaded onto a silica gel column (100–200 mesh). The silica gel column was jacketed so that chromatography could be carried out at 0 °C, and it was eluted with 20% ether in hexane. A 171-mg sample of arachidonic acid (38%) was recovered from this column chromatography along with about 200 mg (45%) of peroxidic materials of TLC fractions A–D (HPETE isomers).

Conversion of the HPETE Isomers into Other Fatty Acid Derivatives. A. HPETE Methyl Esters. A 400-mg sample of *N*-methylnitrosourea[24] was added to 5 mL of 40% KOH and 20 mL of ethyl ether at 0 °C. The mixture was stirred in the dark for 2 h at 0 °C, and the ether layer was removed and then dried over KOH pellets for $^1/_2$ h. This diazomethane/ether solution was then pipetted into a cold (0 °C) ether solution of the HPETE isomers until a faint yellow color persisted. The reaction mixture was then stirred at 0 °C for approximately $^1/_2$ h, and the excess CH_2N_2 was blown off in a hood with a nitrogen stream. The mixture of HPETE methyl esters appeared as four spots: R_f 0.31, 0.25, 0.23, and 0.14.

B. HETE. The HPETE isomer mixture was taken up in ethyl ether that had been previously saturated with water, and a 10% excess of triphenylphosphine was then added. The ether solution was allowed to stand at 0 °C for $^1/_2$ h, and the reaction mixture was then chromatographed on silica gel with ether–hexane (20/80, v/v). The product HETE isomers appeared as four spots upon thin-layer analysis: R_f 0.29, 0.24, 0.21, and 0.04.

C. HETE Methyl Esters. The HPETE isomers were converted to the HPETE methyl esters, and the hydroperoxide group was reduced with triphenylphosphine as described above in A. The HETE methyl esters appear as four spots on thin-layer analysis: R_f 0.59, 0.36, 0.32, and 0.28.

Analysis of Fatty Acid Derivatives as the Methyl (Trimethylsiloxy)eicosanoates. The HPETE and HETE isomers were converted to the methyl esters as described and then converted to the saturated hydroxyeicosanoic methyl esters by catalytic hydrogenation. Ethanol solutions of the substrate to be hydrogenated were allowed to stir for 2 h under a slightly positive hydrogen pressure in the presence of catalytic amounts of platinum oxide. The catalyst was removed by gravity filtration through Celite. The ethanol solvent was removed under vacuum, and the residue was taken up in 100 μL of triethylamine under a nitrogen atmosphere. A 200-μL sample of bis(trimethylsilyl)trifluoroacetamide was added, and the solution was allowed to stir overnight. The solvent was removed in vacuo, and the residue was taken up in 200 μL of pentane and analyzed by gas chromatography–mass spectrometry.

Gas chromatography–mass spectrometry was carried out on a Hewlett-Packard 5992 GC–MS operating with a 3-ft OV 101 column programmed from 200 to 230 °C. The peak eluting at approximately 6 min was due to the methyl (trimethylsiloxy)eicosanoates. The mass spectrum fragments due to fragmentation α to the trimethylsiloxy group are presented in Table III for the various isomeric methyl (trimethylsiloxy)eicosanoates.

Acknowledgment. Support of this research by NIH Grant HL 17921 is acknowledged. We thank Elizabeth Yarbro for invaluable technical assistance and Dr. Marvin Siegel of Burroughs Wellcome Co. for helpful discussions. N.A.P. acknowledges receipt of an NIH RCDA, 1977–1982.

Registry No. 15-HPETE, 69371-38-6; 14-HPETE, 70968-77-3; 12-HPETE, 71030-35-8; 11-HPETE, 70968-78-4; 9-HPETE, 70968-79-5; 8-HPETE, 70968-80-8; 6-HPETE, 70968-81-9; 5-HPETE, 70968-82-0; 15-HPETE methyl ester, 70968-83-1; 14-HPETE methyl ester, 70968-84-2; 12-HPETE methyl ester, 70968-85-3; 11-HPETE methyl ester, 70968-86-4; 9-HPETE methyl ester, 70968-87-5; 8-HPETE methyl ester, 70968-88-6; 6-HPETE methyl ester, 70968-89-7; 5-HPETE methyl ester, 70968-90-0; 15-HETE, 71030-36-9; 14-HETE, 70968-91-1; 12-HETE, 71030-37-0; 11-HETE, 71030-38-1; 9-HETE,

70968-92-2; 8-HETE, 70968-93-3; 6-HETE, 70968-94-4; 5-HETE, 71030-39-2; 15-HETE methyl ester, 71030-40-5; 14-HETE methyl ester, 71000-87-8; 12-HETE methyl ester, 71030-41-6; 11-HETE methyl ester, 70968-95-5; 9-HETE methyl ester, 70968-96-6; 8-HETE methyl ester, 70968-97-7; 6-HETE methyl ester, 70968-98-8; 5-HETE methyl ester, 70968-99-9; methyl 15-(trimethylsiloxy)eicosanoate, 70969-00-5; methyl 14-(trimethylsiloxy)eicosanoate, 70969-01-6; methyl 12-(trimethylsiloxy)eicosanoate, 70969-02-7; methyl 11-(trimethylsiloxy)eicosanoate, 70969-03-8; methyl 9-(trimethylsiloxy)eicosanoate, 70969-04-9; methyl 8-(trimethylsiloxy)eicosanoate, 70969-05-0; methyl 6-(trimethylsiloxy)eicosanoate, 70969-06-1; methyl 5-(trimethylsiloxy)eicosanoate, 70969-07-2; arachidonic acid, 506-32-1.

Ozonolysis of Vinyl Fluoride: Identification of Ozonides and Doubly Fluorinated Products[1]

Ursula Mazur, Robert P. Lattimer, Alexander Lopata, and Robert L. Kuczkowski*

Department of Chemistry, University of Michigan, Ann Arbor, Michigan 48109

Received January 29, 1979

A number of products have been identified upon ozonolysis of vinyl fluoride at low temperatures either neat or in CH_3Cl. The major volatile products are formyl fluoride and vinyl fluoride ozonide (3-fluoro-1,2,4-trioxolane). Small amounts of both cross ozonides have been observed and trace amounts of acetyl fluoride, fluoroacetyl fluoride, and *cis*-1,2-difluorooxirane. The formation of the three ozonides is consistent with a Criegee pathway involving both H_2COO and HFCOO intermediates. The other products can be accounted for by further reactions of intermediates derived from a Criegee pathway or by other unrelated processes. The decomposition of vinyl fluoride ozonide leads to additional volatile products including CO_2, HCOF, HCOOH, SiF_4 (in glass), and two minor compounds identified as bis(fluoromethyl) ether and fluoromethyl formate. The characterization of vinyl fluoride ozonide and fluoromethyl formate ($CH_2FOC(O)H$) by IR, NMR, mass, and microwave spectroscopy is reported.

The condensed phase ozonolysis of low molecular weight alkenes with halogen substitution at the double bond has recently received attention. Low-temperature IR studies[2] on chloroethylenes identified a stable primary ozonide (1,2,3-trioxolane) only for vinyl chloride. No secondary ozonide (1,2,4-trioxolane) formation was observed, unlike a similar study of unhalogenated alkenes.[3]

Griesbaum et al.[4] have examined the ozonolysis of 2,3-dichloro- and 2,3-dibromo-2-butene. Stable ozonides were not isolated. A variety of other products were identified which could be derived from subsequent reactions of the acetyl halide and halogenated carbonyl oxide (HXCOO) expected from a Criegee cleavage[5] of the alkene. However, as much as 30% of the alkene reacted by non-Criegee pathways to produce products such as epoxides or tetrahalobutanes. Ozonolysis of monochloroalkenes in a participating solvent capable of trapping a carbonyl oxide indicated that 80–95% of those alkenes cleaved to give an acyl chloride and carbonyl oxide.[6]

The ozonolysis of perfluoroethylene[7] or 1,2-difluoroethylene[8] has yielded some secondary ozonides. The ozonide yield was small in the case of C_2F_4 where perfluoroethylene oxide and CF_2O were the major volatile products with small amounts of perfluorocyclopropane also observed. The major volatile products from 1,2-$C_2H_2F_2$ are HCOF, the secondary ozonide, and *cis*-1,2-difluorooxirane. A trace amount of *cis*-1,2,3-trifluorocyclopropane was also obtained in methyl chloride solvent.

Such results raise a question as to what extent the normal ozonolysis mechanism[5,9-11] is applicable to halogenated alkenes. We report here our analysis of the ozonolysis of vinyl fluoride.

Results

The ozonolyses of C_2H_3F were conducted at low temperature either in methyl chloride or in isobutane or neat, using standard techniques[9] as described in the Experimental Section. A workup procedure involving distillation of the crude reaction mixture through traps held at −63, −95, and −196 °C and subsequent VPC and spectroscopic analysis of each trap was devised. The coldest trap was found to contain HCOF, C_2H_3F, and (where applicable) solvent. The −95 °C trap after VPC separation was found to contain vinyl fluoride ozonide [3-fluoro-1,2,4-trioxolane (**1**)], both cross ozonides [ethylene ozonide (**2**) and 3,5-difluoro-1,2,4-trioxolane (**3**)], and minor amounts of *cis*-1,2-difluorooxirane (**4**), acetyl fluoride (**5**), and fluoroacetyl fluoride (**6**). Variable amounts of H_2CO and HCOOH were found but they could be reduced to trace amounts in favorable runs. The −63 °C trap contained a clear, unstable liquid which usually generated heat and effervesced upon warming to room temperature. This liquid was predominantly vinyl fluoride ozonide. Its decomposition products were CO_2, HCOF, SiF_4, a brown viscous liquid which needs further characterization, and two minor compounds identified as bis(fluoromethyl) ether [$(CH_2F)_2O$ (**7**)] and fluoromethyl formate [$CH_2FOC(O)H$

(1) (a) A partial report of this work was presented at the Symposium on Ozonolysis, CIC–ACS Meeting, Montreal, June, 1977. (b) Preliminary communication: R. P. Lattimer, U. Mazur, and R. L. Kuczkowski, *J. Am. Chem. Soc.*, **98**, 4012 (1976).

(2) (a) I. C. Hisatsune, L. H. Kolopajlo, and J. Heicklen, *J. Am. Chem. Soc.*, **99**, 3704 (1977); (b) E. Sanhueza, I. C. Hisatsune, and J. Heicklen, *Chem. Rev.*, **76**, 801 (1976).

(3) L. A. Hull, I. C. Hisatsune, and J. Heicklen, *J. Am. Chem. Soc.*, **94**, 4856 (1972).

(4) (a) K. Griesbaum and P. Hofmann, *J. Am. Chem. Soc.*, **98**, 2877 (1976); (b) K. Griesbaum and J. Bruggemann, *Chem. Ber.*, **105**, 3638 (1972).

(5) R. Criegee, *Angew. Chem., Int. Ed. Engl.*, **14**, 745 (1975).

(6) K. Griesbaum and H. Keul, *Angew. Chem.*, **87**, 748 (1975).

(7) F. Gozzo and G. Camaggi, *Chem. Ind. (Milan)*, **20**, 197 (1968).

(8) C. W. Gillies, *J. Am. Chem. Soc.*, **99**, 7239 (1977); **97**, 1276 (1975).

(9) R. P. Lattimer, R. L. Kuczkowski, and C. W. Gillies, *J. Am. Chem. Soc.*, **96**, 348 (1974).

(10) P. S. Bailey and T. M. Ferrell, *J. Am. Chem. Soc.*, **100**, 899 (1978).

(11) V. Ramachandran and R. W. Murray, *J. Am. Chem. Soc.*, **100**, 2197 (1978).

0022-3263/79/1944-3181$01.00/0