Identification of New Geometric Isomers of Methyl Linoleate Hydroperoxide and Their Chromatographic Behavior

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New geometric isomers, methyl (9Z,11Z)-13-hydroperoxy-9,11-octodecadienoate and methyl (10Z,12Z)-9-hydroperoxy-10,12-octodecadienoate, were proved to be present in methyl linoleate hydroperoxide produced by autoxidation. They were identified from their UV, MS, and 1H-NMR spectra after conversion to the corresponding oxo derivatives: methyl (9Z,11Z)-13-oxo-9,11-octodecadienoate and methyl (10Z,12Z)-9-oxo-10,12-octodecadienoate. Their chromatographic behavior is described.

Key words: methyl linoleate; hydroperoxide; autoxidation; oxoocadecadienoate

It is well known that four main isomers of hydroperoxide are produced by the autoxidation of methyl linoleate. These isomers are methyl (9Z,11E)-13-hydroperoxy-9,11-octodecadienoate (OOH 13ZE), methyl (9E,11E)-13-hydroperoxy-9,11-octodecadienoate (OOH 13EE), methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate (OOH 9EZ), and methyl (10E,12E)-9-hydroperoxy-10,12-octodecadienoate (OOH 9EE). In addition to these main isomers, we found that the hydroperoxide produced by autoxidation contained new geometric isomers which had conjugated Z,Z double bonds. This report describes the separation and identification of these isomers after their conversion to the corresponding oxo derivatives (oxo 1 and 2).

Methyl linoleate hydroperoxide used in this experiment was an autoxidation product, its preparation and purification having been described in the previous papers.12 The purified hydroperoxide (7.6 g) was reduced with Pd-C, and the resulting hydroxy derivative was separated by Wako gel C-100 column chromatography, using hexane-diethyl ether (85:15 and 80:20) as the solvent. The separated hydroxy derivative in hexane (5%) was chilled at −80°C to remove the main isomers, methyl (9Z,11E)-13-hydroxy-9,11-octadeicadienoate (OH 13ZE), methyl (9E,11E)-13-hydroxy-9,11-octadecadienoate (OH 13EE), methyl (10E,12Z)-9-hydroxy-10,12-octadecadienoate (OH 9EZ), and methyl (10E,12E)-9-hydroxy-10,12-octadecadienoate (OH 9EE), as a precipitate. This precipitate was filtered off. The hydroxy derivative in the resulting filtrate was converted to oxoocadecadienoate with MnO2 in hexane.3.4 The proportions of oxo 1 and 2 in the total oxo derivative (0.23% and 0.11%) were enhanced to 1.9% and 1.1% by removing the precipitate. Oxo 1 and 2 were separated in a Nucleosil 100-5 column (0.75×25 cm) with hexane-diethyl ether (97:3) at a flow rate of 4.1 ml/min, with monitoring at 268 nm. The retention times of oxo 1 and 2 were 43 min and 52 min, respectively, and oxo 1 and 2 were separated from the four main isomers, methyl (9Z,11E)-13-oxo-9,11-octadecadienoate (oxo 13ZE), methyl (9E,11E)-13-oxo-9,11-octadecadienoate (oxo 13EE), methyl (10E,12Z)-9-oxo-10,12-octadecadienoate (oxo 9EZ), and methyl (10E,12E)-9-oxo-10,12-octadecadienoate (oxo 9EE), which were eluted after 60 min.

The UV absorption maximum of oxo 1 and 2 was λmax 279 nm in ethanol.

Mass spectra were obtained in GC-MS with a JEOL JMS DX-300 mass spectrometer. The GC column (3 mm × 2 m) was packed with Silar 10C, and the column temperature was elevated from 150 to 270°C at a rate of 8°C/min. EI ionization was used with an ionization current of 70 eV. The spectrum of oxo 1 was almost identical to that of...

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Abbreviations: OOH 13ZE, methyl (9Z,11E)-13-hydroperoxy-9,11-octodecadienoate; OOH 13EE, methyl (9E,11E)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate; OOH 9EE, methyl (10E,12E)-9-hydroperoxy-10,12-octodecadienoate; OOH 13EZ, methyl (9E,11Z)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate; OOH 13EZ, methyl (9E,11Z)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate; OOH 13EZ, methyl (9E,11Z)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate; OOH 13EZ, methyl (9E,11Z)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate; OOH 13EZ, methyl (9E,11Z)-13-hydroperoxy-9,11-octodecadienoate; OOH 9EZ, methyl (10E,12Z)-9-hydroperoxy-10,12-octodecadienoate;
methyl (9E,11Z)-13-oxo-9,11-octadecadienoate (oxo 13EZ).\(^1\) The molecular peak was at \(m/z\) 308, and the major fragment peaks were at \(m/z\) 99 [\(\text{CH}_3(\text{CH}_2)_2\text{CO}\)], 71 [\(\text{CH}_3(\text{CH}_2)_4\)], and 151 [\(\text{M}-(\text{CH}_2)_2\text{COOCH}_3\)]. The spectrum of o xo 2 was almost identical to that of methyl (10Z,12E)-9-oxo-10,12-octadecadienoate (oxo 9ZE).\(^1\) The molecular peak appeared at \(m/z\) 308, and the major fragment peaks were at \(m/z\) 277 (M-31), 237 [\(\text{M}-\text{CH}_3(\text{CH}_2)_3\)], 151 [\(\text{M}-(\text{CH}_2)_2\text{COOCH}_3\)], and 185 [\(\text{CO}(\text{CH}_2)_2\text{COOCH}_3\)]. Oxo 1 and 2 were reduced with NaBH\(_4\) and hydrogenated with platinum-black to determine the positions of the o xo groups. The mass spectrum of the TMS derivative of reduced o xo 1 showed strong fragment peaks at \(m/z\) 173 and 315,\(^6\) which indicated that the position of the TMSO group was at C-13. The mass spectrum of the TMS derivative of reduced o xo 2 showed \(m/z\) 259 and 229,\(^6\) which indicated that the TMSO group was at C-9.

\(^1\)H-NMR spectra were obtained with a Bruker AMX 400WB spectrometer (400 MHz) in CDCl\(_3\) with TMS as an internal standard. The signals of four olefinic protons appeared at \(\delta\) 5.8–7.2 ppm. In o xo 1, a signal at \(\delta\) 5.94 (doublet) was assigned to H-12, and in o xo 2, a signal at \(\delta\) 5.93 was assigned to H-10 from their coupling patterns. H-H COSY spectra were determined to assign the signals at \(\delta\) 5.84, 6.68, and 7.21 in o xo 1, and at \(\delta\) 5.86, 6.70, and 7.20 in o xo 2. In o xo 1, the signal at \(\delta\) 5.84 was coupled with the signals at \(\delta\) 7.21 and 2.19 (a methylene proton adjacent to an olefin), so this was assigned to H-9. The signal at \(\delta\) 6.68 was coupled with the signals at \(\delta\) 7.21 and 5.94 (H-12), and the signal at \(\delta\) 7.21 was coupled with the signals at \(\delta\) 5.84 (H-9) and 6.68. Consequently, the signal at \(\delta\) 6.68 was assigned to H-11, and that at \(\delta\) 7.21 was assigned to H-10. Similarly, in o xo 2, the signal at \(\delta\) 5.86 was assigned to H-13, that at \(\delta\) 6.70 to H-11, and that at \(\delta\) 7.20 to H-12. The geometry of the double bond of o xo 1 and 2 was determined by the coupling constants. In o xo 1, \(J_{11,12}\) was 11.4 Hz and \(J_{9,10}\) was 11.0 Hz, and in o xo 2, \(J_{10,11}\) = 11.5 Hz and \(J_{12,13}\) was 11.0 Hz, which indicates that both of the two double bonds are of Z configuration.\(^{1,5}\)

From the UV,\(^6\) MS, and \(^1\)H-NMR results, o xo 1 and 2 were identified as methyl (9Z,11Z)-13-oxo-9,11-octadecadienoate and methyl (10Z,12Z)-9-oxo-10,12-octadecadienoate.

O xo 1 and 2 originated from the corresponding isomers of the hydroperoxide of methyl linoleate, and the chromatographic behavior of these new geometric isomers of hydroperoxide (OOH 1 and 2) was examined. The hydroperoxide of methyl linoleate gave four peaks by HPLC and was separated into 8 fractions as shown in Fig. 1. The four peaks in Fig. 1 are OOH 13EZ, OOH 13EE, OOH 9EZ, and OOH 9EE, which were determined as described in the previous paper.\(^{1,5}\) The separated fractions were treated with \(\phi_3\)P and with MnO\(_2\), and the concentrations of o xo 1 and 2 in each fraction were determined by HPLC. O xo 1 was detected mainly in fractions 3 and 4 (22.5% and 65.6% of total o xo 1), and o xo 2 mainly in fraction 8 (91.8% of total o xo 2). Accordingly, OOH 1 was eluted together with OOH 13EE, and OOH 2 was eluted together with OOH 9EE.

O xo 1 and 2 were reduced with NaBH\(_4\), and the retention times of the corresponding hydroxy derivatives were compared with those of the main isomers (OH 13EZ, OH 13EE, OH 9EZ, and OH 9EE). OH 1 was eluted between OH 13EE and OH 9EZ, and OH 2 was eluted after OH 9EE.

Thus, for all of the geometric isomers that have hydroperoxy, hydroxy, and o xo groups at C-9 and C-13, the chromatographic behavior was established as in the previous paper\(^{1,5}\) and the present investigation. The chromatographic behavior of each geometric isomer by HPLC is schematically shown in Fig. 2. All 8 isomers in the o xo derivatives can be resolved. Isolation of the minor isomers is easy because the minor isomers (o xo 1, o xo 2, o xo 13EZ, and o xo 9ZE) are eluted more rapidly than the four main isomers. In the hydroperoxide and hydroxy derivatives, the minor isomers are eluted near the main isomers. In the hydroxy derivatives, although OH 1 and 2 can be separated from the main isomers, they give no peak when a hydroxy derivative from an autoxidation product is analyzed by HPLC because the contents of OH 1 and 2 are very low. In the hydroperoxide, the minor methyl (9E,11Z)-13-hydroperoxy-9,11-octadecadienoate (OOH 13EZ) and methyl (10Z,12Z)-9-hydroperoxy-10,12-octadecadienoate (OOH 9ZE) isomers can be sepa-
Fig. 2. Summarized Chromatographic Behavior of Each Isomer of the Hydroperoxide (ROOH), Hydroxy Derivative (ROH), and Oxo Derivative (R = O).


rated from the main isomers, although their peaks are often obscure.

Porter et al. have reported⁷ that conjugated hydroperoxides other than the 4 known isomers could be detected when linoleate was co-oxidized with a high concentration (1.5–3 m) of cyclohexadiene. Although they did not identify these hydroperoxides, they considered that they were isomers geometrically different from the known ones. They also considered that these hydroperoxides were generated from isomerized pentadienyl radicals. These isomers are provably the same as those identified in our previous work⁷ and the present paper.

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References


