

α - AND β -MYRCENE HYDROPEROXIDE FROM *ARTEMISIA ANNUA*

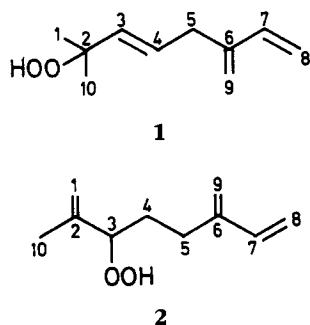
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Recently, two sesquiterpene peroxides were isolated from the aerial parts of *Artemisia annua* L. (Asteraceae), qinghaosu (1-3), and artemisitene (4). Qinghaosu is used as a potent antimalarial drug in China (2).

After chromatographic purification of the crude extract of *A. annua* by repeated column chromatography, two further compounds [1,2] were obtained. Both are very unstable and give positive reactions with NaI-starch reagent.

The ms of compound 1 shows a very



weak molecular ion at m/z 168 (0.4%). The exact mass measurement (m/z 168.1126) corresponds to the molecular formula $C_{10}H_{16}O_2$ (required 168.1150), indicating three double bond equivalents. The ir shows bands for a vinyl group at 990 and 900 cm^{-1} as well as for a conjugated diene at 1665 and 1590 cm^{-1} , which is also indicated by the uv maximum at 223 nm. The double band at 1380 cm^{-1} suggests a geminal dimethyl group. The sharp OH stretching vibration at 3550 cm^{-1} and the signal of one proton in the 1H nmr at 7.28 (s) ppm, which can be exchanged by D_2O , marks a hydroperoxy group. The 1H nmr (Table 1) exhibits seven olefinic protons from 4.99-6.50 ppm, including the ABX system of a vinyl group at 6.38, 5.23, and 5.08 ppm as well as two *trans*-positioned protons at 5.78 (dt) and 5.60 (dt) ppm. In addition, the doublet of a CH_2 -group at 2.97 ppm and a singlet at 1.33 ppm for two magnetically equivalent methyl groups appear.

TABLE 1. 1H -nmr Data for Compounds 1 and 2 ($CDCl_3$)^a

Proton	Compounds	
	1	2
H-1	1.33 s	5.05 s (br) ^c 5.02 s (br) ^c
H-3	5.60 dt ($J_{3,4}$ 16; $J_{3,5}$ 1)	4.37 t ($J_{3,4}$ 7)
H-4	5.78 dt ($J_{4,3}$ 16; $J_{4,5}$ 6.5)	1.7 m
H-5	2.97 (br) ($J_{5,4}$ 6.5)	2.28 m
H-7	6.38 dd ($J_{7,8'}$ 17.5; $J_{7,8}$ 10.2)	6.34 dd ($J_{7,8'}$ 17.5; $J_{7,8}$ 10.5)
H-8	5.08 dd ($J_{8,8'}$ 1.3; $J_{8,7}$ 10.2)	5.08 dd ($J_{8,7}$ 10.5)
H-8'	5.23 d ($J_{8',8}$ 1.3; $J_{8',7}$ 17.5)	5.25 dd ($J_{8',7}$ 17.5)
H-9	5.06 bs ^b	5.05 s (br) ^d
H-9'	4.99 t	5.02 s (br) ^d
H-10	1.33 s	1.75 s
OOH	7.28 s	7.82 s

^aShifts in ppm and coupling constants as Hz.

^{b,c,d}Overlapping.

In agreement with the ^{13}C -nmr data (Table 2) which are assigned by comparison with the ^{13}C -nmr data of β -myrcene, the structure of 2-hydroperoxy-2-methyl-6-methylene-3,7-octadiene (β -myrcene hydroperoxide) results for **1**.

TABLE 2. ^{13}C -nmr Data for Compounds **1** and **2** (CDCl_3)^a

Catom	Compounds	
	1	2
C-1	23.9	114.1
C-2	81.9	145.2
C-3	134.8	88.9
C-4	128.9	29.1
C-5	34.1	27.0
C-6	144.2	143.2
C-7	138.2	138.3
C-8	116.6	115.8
C-9	113.5	113.1
C-10	23.9	16.98

^aProton broad band decoupling.

For the extremely unstable compound **2**, a molecular ion was not detectable. The ^1H - and ^{13}C -nmr spectra (Tables 1,2) suggest a monoterpene with the molecular formula $\text{C}_{10}\text{H}_{16}\text{O}_2$. In the ir bands for double bonds are present at 3085, 1740, 1640, 990, and 900 cm^{-1} . The HO stretching vibration at 3540 cm^{-1} as well as a signal in the ^1H nmr at 7.82 (s) ppm disappearing upon D_2O addition, indicate a hydroperoxy group. In the ^1H nmr (Table 1) there are signals for seven olefinic protons within the range of 5.00-6.24 ppm, including the ABX-system of a vinyl group at 6.34, 5.25, and 5.08 ppm, resembling the ^1H nmr of **1**. Furthermore, the triplet of a carbinol proton at 4.37 ppm marks a secondary hydroperoxy group (5), and there are multiplets of two methylene groups at 2.28 and 1.70 ppm as well as the singlet of an isolated methyl group at 1.75 ppm. In accordance with the ^{13}C nmr (Table 2) which has been assigned by comparison with β -myrcene, compound **2** has the structure of 3-hydroperoxy-2-methyl-6-methylene-1,

7-octadiene (α -myrcene hydroperoxide). Compounds **1** and **2** have previously been synthesized by the ene-reaction of β -myrcene (6,7). The isolated compounds, which were found for the first time in nature, are identical in their ir, ^1H -nmr, and ^{13}C -nmr spectra with the compounds prepared from β -myrcene according to the literature (6,7).

EXPERIMENTAL

Ir spectra were obtained with a Perkin-Elmer 298 spectrometer in CCl_4 . ^1H -nmr and ^{13}C -nmr spectra were recorded on a Varian XL 300 Spectrometer in CDCl_3 . The mass spectrum was obtained with a Kratos MS 50 mass spectrometer. Uv spectra were run in cyclohexane on a Perkin-Elmer Model 550S.

The seeds of *A. annua* were delivered from botanic gardens of the universities of Frankfurt/Main, Stuttgart/Hohenheim, and from Southern Europe. They were planted in a greenhouse and were then cultivated outdoors. The plants were harvested during their blossom time at the beginning of October 1985. A voucher specimen of the plant is preserved at the Herbarium of the Botanical Institute, University of Bonn.

2-HYDROPEROXY-2-METHYL-6-METHYLENE-3,7-OCTADIENE (β -MYRCENE HYDROPEROXIDE) [1]—Fresh, crushed aerial parts of *A. annua* (4.5 kg) were extracted with petroleum ether (bp 40-60°) and processed according to Rucker *et al.* (3) to yield approximately 20 g of a viscous, dark green extract. This extract was chromatographed with CHCl_3 -EtOAc (92.5:7.5) on 1950 g Si gel (0.06-0.2 mesh). Two peroxide-containing fractions were collected: Rf 0.44-0.48 (0.16 g) and Rf 0.50-0.56 (1.75 g).

The fraction with Rf 0.44-0.48 was separated on two Lobar-A-columns (Merck, hexane-Et₂O, 85:15), yielding 60 mg **1** (0.3% of the extract) as a colorless, highly volatile, viscous oil: Rf 0.16 in hexane-Et₂O (85:15); ir (CCl_4) 3550, 3090, 2980, 2920, 1740, 1665, 1635, 1590, 1380, 1360, 890 cm^{-1} ; uv (cyclohexane) λ max 223 nm; ms (70 eV, direct inlet at 180°) m/z (%) 168 (M^+ , 0.4), 153(1.4), 151(3.2), 150(1.4), 135(61.5), 119(11.1), 107(29.8), 93(48.8), 91(29.4), 81(17.0), 79(38.5), 77(20.2), 69(23.0), 67(22.6), 59(26.2), 55(35.7), 43(100); ^1H nmr (CDCl_3 , 300 MHz) Table 1; ^{13}C nmr (CDCl_3 , 75.43 MHz) Table 2.

3-HYDROPEROXY-2-METHYL-6-METHYLENE-1,7-OCTADIENE (α -MYRCENE HYDROPEROXIDE) [2]—The fractions with Rf 0.50-0.56 (see **1**) were separated on two Lobar-C-columns (Merck) with hexane-Et₂O (8:2). By preparative tlc (Si gel) with the same solvent, 8 mg (0.04% of the

extract) of a colorless, highly volatile oil was obtained [Rf 0.28 in hexane-Et₂O (8:2)]; ir (CCl₄) 3540, 3085, 2930, 1740, 1645, 1590, 990, 900 cm⁻¹; uv (cyclohexane) λ max 223 nm; ¹H nmr (CDCl₃, 300 MHz) Table 1; ¹³C nmr (CDCl₃, 75.43 MHz) Table 2.

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