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A QUINONE METHIDE FROM *SALVIA OFFICINALIS*

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Key Word Index—*Salvia officinalis*; Labiatae; diterpene; sagequinone methide A.

Abstract—A novel diterpene, sagequinone methide A, was isolated from the ethyl acetate extract of sage, *Salvia officinalis*, whereas no sagequinone methide A could be detected from the methanol extract. Another novel compound, 6,7-dimethoxyrosmanol was isolated from the methanol extract.

INTRODUCTION

Many diterpenes have been isolated from plants in the genus *Salvia* [1–5] and some of them are reported to have potent antioxidative activity [6]. The methyl ethers or the methyl esters of the diterpenes have been also isolated from the methanol extracts of those plants [7]. This suggested to us that some of the reported diterpenes might be artefacts formed during the extraction or the isolation. We examined the diterpenes from the ethyl acetate extract of *S. officinalis* to identify the natural constituents. This led to the isolation of a novel *p*-quinone methide, sagequinone methide A (**1**), which was assumed to be an intermediate in the autooxidation of carnosic acid (**2**) to form rosmanol (**3**) [8]. A novel compound, 6,7-dimethoxy-7-*epi*-rosmanol (**4**), and 7-methoxyrosmanol (**8**) which may be formed from **1** [9], and other diterpenes were isolated from the methanol extract.

RESULTS AND DISCUSSION

The aerial parts (1 kg) of sage, *Salvia officialis* were extracted with ethyl acetate for 20 days. The ethyl acetate-soluble residue was partitioned between ethyl acetate and water. The organic layer was evaporated and the residue was partitioned between hexane and acetonitrile. The hexane-soluble fraction was subjected to silica gel column chromatography and HPLC (TSK gel and Asahipack) to yield compound **1**, carnosic acid (**2**) [6] and methyl carnosoate [6]. Sageone [5, 9], safficinolide [5], carnosol [6], 7-methoxyrosmanol [11], isorosmanol [10], rosmanol [6] and epirosmanol [6, 10] were obtained from acetonitrile-soluble fraction. The known compounds were identified by comparison of their ^{1}H and ^{13}C NMR spectral data with the published data.

Compound **1** was assigned the molecular formula $C_{20}H_{24}O_4$. The presence of a hydroxyl group, a γ-lactone ring system (δ_c 176.2, 72.9), a highly conjugated carbonyl (δ_c 180.4, 143.8, 134.8, 145.8, 133.8, 139.9) and an isopropyl group [δ_H 1.13 (6H, *d*, J = 8.1 Hz), 3.03 (1H, *sept*, J = 8.1 Hz); δ_c 26.9, 21.5, 21.4] were indicated by the IR (ν_{max}^{KBr} cm^{-1}: 3410, 2920, 1760, 1610, 1565), UV [λ_{max}^{EtOH} nm (ε): 287 (2164)], and ^{13}C and ^{1}H NMR data. NMR experiments (H–H COSY, C–H COSY, NOESY) showed the partial structure depicted in Fig. 1. HMBC of **1** showed the connectivities of the partial structures as in Fig. 2. The structure of **1** was thus deduced as 11-hydroxy-12-oxo-7,9(11),13-abietatrieno-20,6-lactone and named sagequinone methide A. Although the formation of sagequinone methide A as an intermediate in the autooxidation of carnosic acid (**2**) to form rosmanol (**3**), was assumed by Wenkert [8], the isolation of quinone methide A has not been reported before. Com-

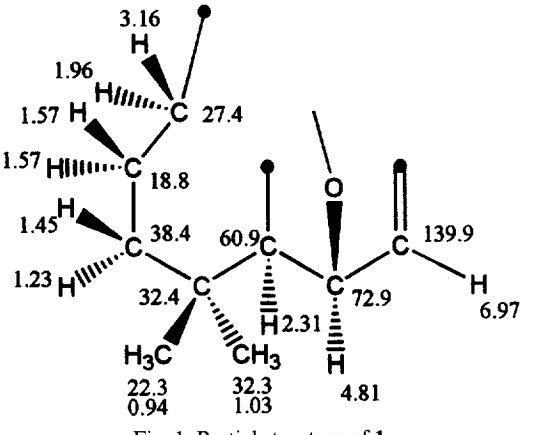

Fig. 1. Partial structure of **1**.

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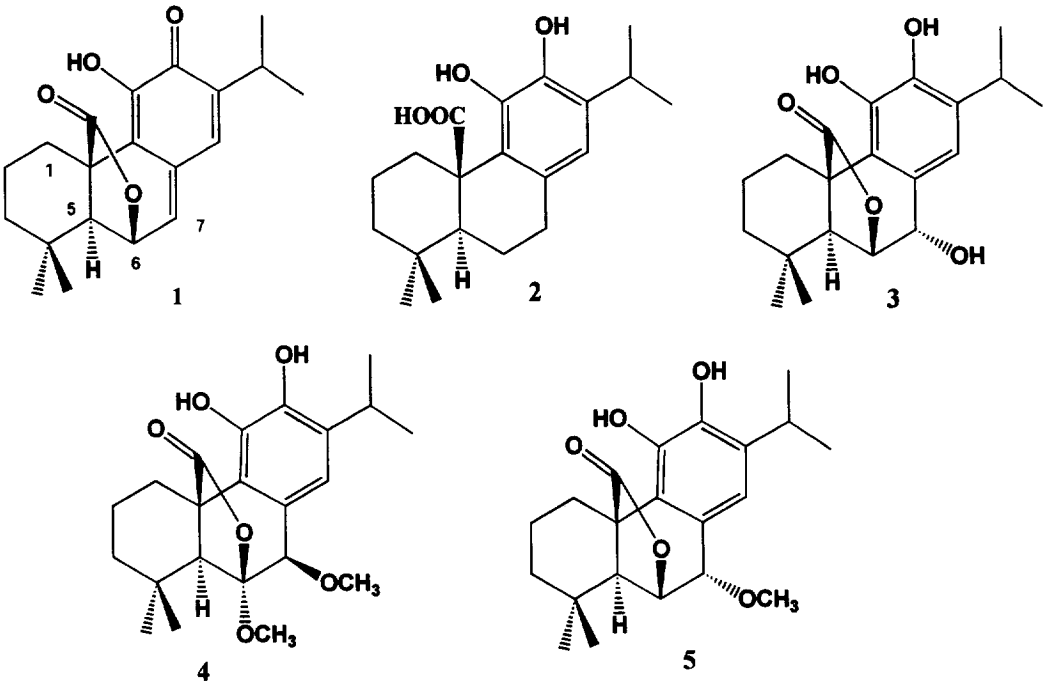

pound **1** was not detected in the methanol extract of sage, whereas a large quantity of 7-methoxyrosmanol was obtained. This suggests that most of the 7-methoxyrosmanol (**5**) is an artefact due to the extraction of the plants with methanol. In fact, sagequinone methide A **1** was converted to 7-methoxyrosmanol **5** in methanol within 2 days at room temperature. From the methanol extract of sage, a novel compound **4** was separated, together with sageone, safficinolide, methyl carnosoate, carnosol, 7-methoxyrosmanol, 12-*O*-methylcarnosic acid and manool.

Compound **4** was assigned the molecular formula of $C_{22}H_{30}O_6$. The spectral data showed the presence of two methoxyl groups [δ_H 3.45 (3H, *s*), 3.32 (3H, *s*), δ_c 53.7, 49.8], an isopropyl group [δ_H 3.02 (1H, *sept*, J = 7.0 Hz), 1.18 (3H, *d*, J = 7.0 Hz), 1.09 (3H, *d*, J = 7.0 Hz), δ_c 27.0, 22.0 22.8], two hydroxyl groups [δ_H 5.97 (1H, *br s*) and 5.82 (1H, *br s*): exchangeable with D_2O, δ_c 142.9, 142.1], an aromatic ring [δ_G 6.93 (1H, *s*)], a γ-lactone ring [ν_{max}^{film} cm^{-1}: 1760 cm^{-1}; δ_c 178.6] and an acetal carbon [δ_c 97.3]. These partial structures and the structures of other constituents from sage indicated that **2** had the abietane skeleton. The NOE between H-5 and H-7 indicated a 7β-OMe as shown in Fig. 3. These confirmed that **2** is 6,7-dimethoxy-7-*epi*-rosmanol which must be an artefact formed from a possible precursor, e.g. isogaldosol [9], during the extraction of the plant with methanol.

EXPERIMENTAL

Mps: uncorr.; ^1H and ^{13}C NMR: 270 and 68 MHz, respectively, $CDCl_3$, TMS as int. standard; CC: silica gel 60 (Merck, 70–230 mesh) with hexane and EtOAc; HPLC: ODS column (LiChrospher RP-18 and Inertsil PREP-ODS, eluted with MeOH–H_2O) and gel per-

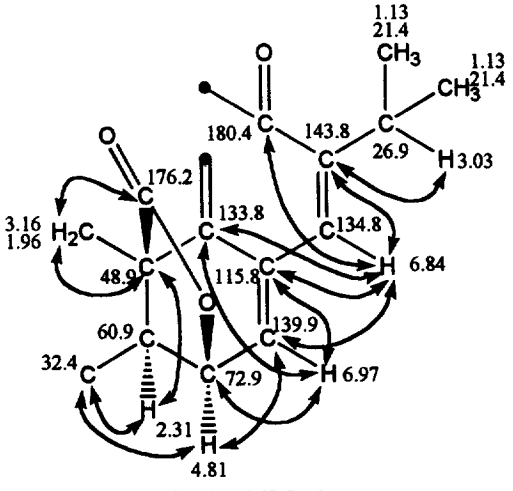

Fig. 2. HMBC of **1**.

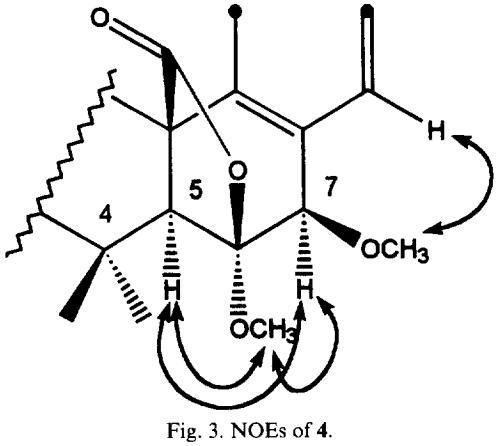

Fig. 3. NOEs of **4**.

meation column (TSK gel G-1000H 6, eluted with $CHCl_3$ and Asahipak GS310, eluted with EtOAc) linked to UV detector.

Plant material. Salvia officinalis was harvested in Dalmatia, late Yugoslavia in 1988.

Extraction and isolation. Dry leaves and stems (1.0 kg) were extracted with EtOAc at room temp. for 20 days. The extract was concd *in vacuo* and the residue partitioned between EtOAc and H_2O. The organic layer was evapd and the residue was partitioned between hexane and MeCN. The hexane-soluble fr. was subjected to silica gel CC eluted with hexane–EtOAc and then HPLC (TSK gel, eluted with $CHCl_3$ and Asahipack, eluted with EtOAc) to afford compound **1** (207 mg), carnosic acid (353 mg) and methyl carnosoate (452 mg). The MeCN-soluble fr. was purified by the similar procedures described for the hexane-soluble fr. to yield sageone (22 mg), safficinolide (57 mg), carnosol (337 mg), 7-methoxyrosmanol (2 mg), isorosmanol (57 mg), rosmanol (113 mg) and epirosmanol (1 mg). The structures of the known compounds were identified by comparison of their NMR spectral data (^1H and ^{13}C) with the published data.

The MeOH extract of sage (1.15 kg) was evapd and the residue was partitioned between EtOAc and H_2O. The EtOAc-soluble fr. was subjected to silica gel CC and then HPLC (TSK gel, eluted with $CHCl_3$ and Asahipack, eluted with EtOAc) to give **4** (2 mg), 12-*O*-methylcarnosic acid (19.1 mg) and manool (1.7 g) together with the abietanes described in our previous report [5].

Sagequinone methide A (**1**). Yellow crystal, mp 145–147°, $[\alpha]_D^{25} -80.69°$ (*c* 0.29, $CHCl_3$); IR ν_{max}^{KBr} cm^{-1}: 3410, 2920, 1760, 1610; UV λ_{max}^{EtOH} (log ε): 287 (2164); HRMS (Found: M^+, 328.1698. $C_{20}H_{24}O_4$ requires M, 328.1674); ^1H NMR: δ 7.64 (1H, *s*), 6.97 (1H, *d*, $J = 2.7$ Hz), 6.84 (1H, *s*), 4.81 (1H, *d*, $J = 2.7$ Hz), 3.16 (1H, *m*), 3.03 (1H, *sept*, $J = 8.1$ Hz), 2.31 (1H, *s*), 1.96 (1H, *ddd*, $J = 13.5, 13.5, 5.1$ Hz), 1.57 (2H, *m*), 1.45 (1H, *br.d*, $J = 13.5$ Hz), 1.23 (1H, *dd*, $J = 13.5, 6.1$ Hz), 1.13 (6H, *d*, $J = 8.1$ Hz), 1.03 (3H, *s*), 0.94 (3H, *s*); ^{13}C NMR: δ 180.4, 176.2, 145.8, 143.8, 139.9, 134.8, 133.8, 115.8, 72.9, 60.9, 48.9, 38.4, 32.4, 27.4, 26.9, 22.3, 21.5, 21.4, 18.8.

6,7-*Dimethoxy*-7-epi-*rosmanol* (**4**). Yellow crystal, mp 183–188°, $[\alpha]_D^{25} -30.0°$ (*c* 0.02, $CHCl_3$); IR ν_{max}^{film} cm^{-1}: 3460, 2950, 1760; UV λ_{max}^{EtOH} nm (log ε): 288 (5331); HRMS (Found: M^+, 390.2038. $C_{22}H_{30}O_6$ requires M, 390.2043); ^1H NMR: δ 6.93 (1H, *s*), 5.97 (1H, *br s*)*, 5.82 (1H, *br s*)*, 4.68 (1H, *s*), 3.35 (3H, *s*), 3.32 (3H, *s*), 3.19 (1H, *m*), 3.02 (1H, *sept*, $J = 7.0$ Hz), 2.40 (1H, *s*), 2.01 (1H, 2.01, *ddd*, $J = 14.1, 14.1, 6.1$ Hz), 1.3–1.8 (4H), 1.18 (3H, *d*, $J = 7.0$ Hz), 1.09 (3H, *d*, $J = 7.0$ Hz), 1.03 (3H, *s*), 0.97 (3H, *s*); *: exchangeable with D_2O]; ^{13}C NMR: δ 178.6, 142.9, 142.1, 133.9, 125.9, 122.4, 118.7, 97.3, 75.8, 53.7, 49.8, 49.2, 48.2, 38.4, 31.7, 31.4, 27.3, 27.0, 22.8, 22.0, 21.9, 19.0.

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