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GERMACRANOLIDES AND A MONOTERPENE CYCLIC PEROXIDE FROM *ARTEMISIA FRAGRANS*

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Key Word Index—*Artemisia fragrans*; Compositae; Anthemideae; Seriphidium; sesquiterpene lactones; germacranolides; cyclic monoterpene peroxide.

Abstract—The aerial parts of *Artemisia fragrans* collected in Armenia yielded a new cyclic monoterpene peroxide with the irregular santolinyl framework, together with several known germacranolides. Comparison with previously published chemical results suggests these may actually have been performed on a different, although closely related species.

INTRODUCTION

The large genus *Artemisia* has been the object of numerous chemical studies [1]. In continuation of our studies on this topic, we have investigated *A. fragrans* Willd. [2, 3], a species which grows in Russia, Armenia and neighbouring domains. Previous chemical investigations reported the isolation of several sesquiterpene lactones with germacrane, eudesmane, guaiane and elemane frameworks [4 and references therein]. The structures proposed for shonachalins A–D were shown to be totally or partially incorrect and alternative structures proposed [5–8]. For this reason, we became interested in isolating these lactones from the aforementioned species in order to prove our assumptions in a definitive way.

RESULTS AND DISCUSSION

The aerial parts of *A. fragrans* collected in Armenia gave known germacranolides **1**–**8** [8], the eudesmanolide taurin [9], and the monoterpenes **9** [5] and **10**, the latter being a new compound.

The IR (Experimental) and NMR data (Table 1) of compound **10**, $C_{12}H_{20}O_5$, isolated in a very small amount, indicated the presence of a primary hydroxyl and an acetate group. Furthermore, one C=C bond was also present. With the aid of decoupling experiments, the fragments —CH(OR)—CH(C_4)—CH(OR)—CH_2OH, CH_2=C(CH_3) and (CH_3)$_2$C—OR were established. These fragments were connected to structure **10** after consideration of the heteronuclear long-range correlations (HMBC). Like **9**, compound **10** displays an irregular monoterpene framework of the santolinyl type [10]. An alternative oxetane structure was rejected on the basis of its calculated J values, which were very different from the observed ones. The relative configuration has been proposed on the basis of coupling constant values and NOE measurements. Figure 1 contains a perspective of the optimized geometry of the molecule as calculated by PCMODEL [11].

In the formulae **1**–**10**, we have arbitrarily drawn compound **10** with the same absolute configuration at C-4 as its most likely biosynthetic precursor, the naturally occurring monoterpene (*S*)-(+)-santolina triene [10]. This compound is widespread in many essential oils, especially in the tribe Anthemideae and in the genus *Artemisia* [1]. Monoterpene **10**, as well as **9**, may be biogenetically formed from santolina triene by sequential oxidative modifications of the C=C bonds. A fact which lends support to this biogenetic proposal is the occurrence in other *Artemisia* species of hydroperoxides formed most likely by enzymatic oxygenation of santolina triene [12]. Metabolites of this type are also common in other members of the tribe Anthemideae [13].

It is noteworthy that the main chemical components of *A. fragrans* found by us are essentially coincident with those of North African *A. herba-alba* [8]. The characteristic 9-oxygenated *trans*-12,6-germacranolides, together with 9-oxygenated *trans*-12,6-eudesmanolides, have been reported within the genus

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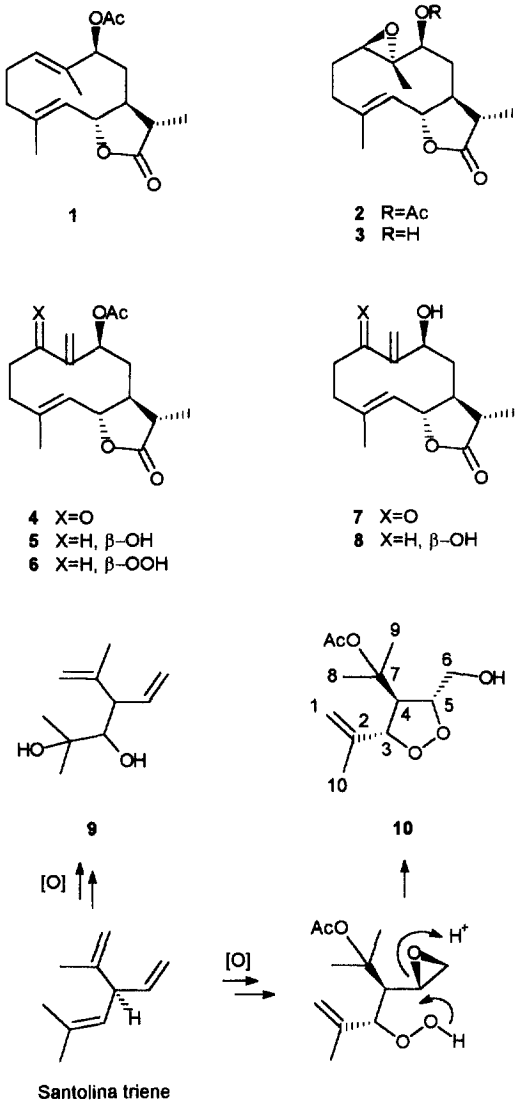

only in the latter species and in *A. vallesiaca* [14]. The results described here and those previously published on a putative *A. fragrans* [4] differ completely. These differences suggest that the previous investigations

Table 1. ^{1}H and ^{13}C NMR data of monoterpene peroxide **10**

H		C	
1	5.18 *br s*	1	118.1
	5.08 *quint* (1.5)	2	139.8
3	4.60 *d* (7)	3	86.6
4	3.10 *dd* (7; 3.8)	4	60.4
5	4.31 *ddd* (8; 3.8; 3)	5	84.2
6	3.77 *br dd* (12; 8)	6	63.4
	3.57 *br d* (12)	7	81.6
8/9	1.56 *s*/1.52 *s*	8/9	24.1/24.0
10	1.77 *br s*	10	17.3
OAc	1.95 *s* (3H)	OAc	170.1/22.2
OH	2.05 *br s*		

δ in ppm and J (parentheses) in Hz.

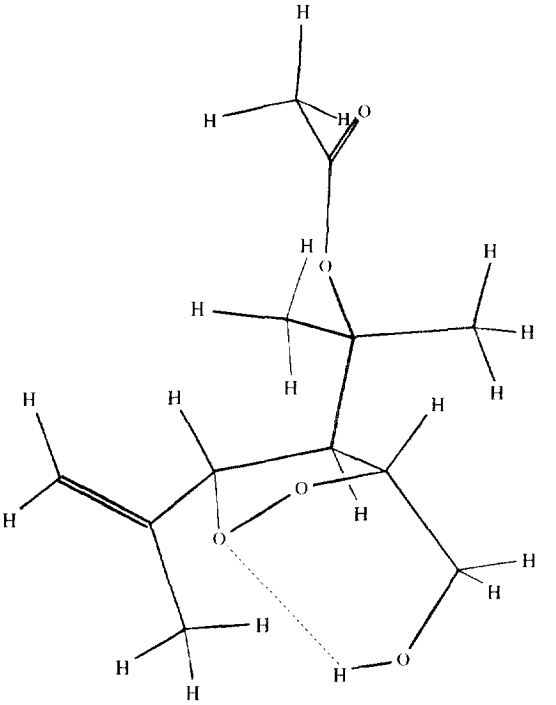

Fig. 1. Optimized geometry of peroxide **10** (PCMODEL).

may not have actually been performed on *A. fragrans* but rather on a closely related species. This related species might be, for instance, *A. oliveriana* [2], which grows in the same geographical zones as *A. fragrans* (in a recent disclosure, *A. fragrans* has been considered to be synonymous with *A. erivanica* (*A. fragrans* var. *erivanica*) and *A. maritima* var. *erivanica* [15]). This assumption is suggested by the fact that two compounds found by us in *A. oliveriana* [7] are in all probability identical with shonachalins A and C, two of the sesquiterpene lactones reported to occur in *A. fragrans* [4].

EXPERIMENTAL

NMR: 400 MHz (^{1}H) and 100 MHz (^{13}C) in $CDCl_3$ (22°). The solvent signals were used as the reference. NOE measurements were carried out by the one-dimensional difference method. OR: 22°. CC: silica gel Merck (particle size 50–200 μ), gradient elution with the solvent mixts indicated in each case. HPLC: LiChrosorb RP-8 (250 × 8 mm), elution with MeOH–H_2O mixts.

Plant material. Aerial parts of *A. fragrans* were collected in the Ardanish Peninsula, province of Sevan, Armenia, at 2000 m. altitude on 17 August 1995. A voucher specimen (BCF-42121) has been deposited in the herbarium of the Laboratory of Botany, Faculty of Pharmacy, University of Barcelona, Spain (Prof. J. Vallès-Xirau).

Extraction and chromatography. The plant material was processed according to the described protocol [16]. The defatted extract was prefractionated by CC on silica gel (fr. A, hexane–EtOAc 3:1; fr. B, hexane–

EtOAc 1:3; fr. C, EtOAc–MeOH 9:1). The three frs were subjected to further chromatographic sepns as described below. The compounds were eluted in the indicated order, which corresponds to increasing polarity on normal-phase silica gel.

From fr. A, after CC on silica gel (elution with hexane–*t*BuOMe 1:1→*t*BuOMe) and, where necessary, prep. TLC, the following compounds were isolated: taurin (9 mg), **1** (263 mg), **9** (157 mg), **10** (4 mg), **4** (18 mg), **2** (34 mg) and **5** (57 mg).

CC of fr. B (CH_2Cl_2–EtOAc 12:1→EtOAc), followed where necessary by prep. TLC, allowed the isolation of **1** (27 mg), **4** (54 mg), **2** (100 mg), **6** (295 mg), **5** (410 mg), **3** (7 mg) and **8** (13 mg).

Fr. C (CC with CH_2Cl_2–EtOAc–MeOH 20:5:1→5:5:1→EtOAc–MeOH 5:1), followed where necessary by prep. TLC and/or HPLC afforded **5** (113 mg), **7** (48 mg) and **8** (390 mg).

(3S*,4R*5R*)-3-*Hydroxymethyl*-4-(1-*acetoxyisopropyl*)-5-*isopropenyl*-1,2-*dioxolane* (**10**). *Oil*, $[\alpha]_D$ −11.5° ($CHCl_3$; *c* 0.7); IR ν_{max}^{film} cm^{-1}: 3440 (*br*, OH), 1727 (acetate C=O), 1451, 1376, 1252, 1141, 1032, 732; EIMS (probe) *m/z* (rel. int.): 203 $[M—C_3H_5]^+$ (10), 125 (24), 109 (36), 83 (100), 69 (80), 55 (41); NMR: Table 1.

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