(*t*), 27.4 (*t*), 35.9 (*t*), 37.7 (*d*), 37.9 (*d*), 38.7 (*t*), 45.3 (*t*), 49.2 (*s*), 85.4 (*s*), 121.5 (*d*), 139.7 (*s*).

Acknowledgements—We are grateful to Dr. J. Schmidt, Institute of Plant Biochemistry, Halle, for his help in collecting the plant material and for performing the GC-MS analysis of the sterols, to Professor Y. Asakawa (Institute of Pharmacognosy, Tokushima) for analysis of the mixture of sesquiterpenes and to Dr. I. H. Sadler and the SERC Highfield NMR Service, University of Edinburgh.

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SESQUITERPENE LACTONES FROM *LASERPITIUM GARGANICUM*

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Key Word Index—*Laserpitium garganicum* subsp. *garganicum*; Umbelliferae; sesquiterpene lactones.

Abstract—Besides known compounds, the roots of *Laserpitium garganicum* subsp. *garganicum* afforded a new slovanolide triester. The locations of the ester moieties were assessed by correlation with a compound of known structure.

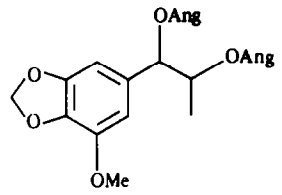
INTRODUCTION

The genus *Laserpitium* is a rich source of oxygenated sesquiterpenoids [1]. As a part of an investigation on plants of this genus, we have studied the constituents of *L. garganicum* (Ten.) Bertol., a species confined to the Italian peninsula and Sardinia (*L. garganicum* subsp. *garganicum*), Sicily [*L. garganicum* subsp. *siculum* (Sprengel) Pign.] and possibly the Balkans [2].

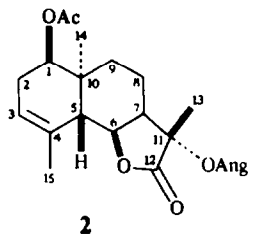
RESULTS AND DISCUSSION

Column chromatography of the chloroform extract of the roots of *L. garganicum* subsp. *garganicum* gave, along with the phenylpropane derivative laserin (**1**) [3], five sesquiterpene lactones (**2**–**6**). Lactone **2** was the eudesmanolide isosilerolide [4]. The other lactones were guaiane-type esters based on the slovanolide skeleton [5]. They differed from each other only in the nature and locations of the ester moieties, as shown by their conversion to the same rearranged triol (**7**) upon treatment with methanolic KOH [6]. Lactones **4**–**6** were identified as guaianolides previously isolated from *L. siler* L. [5]. However, lactone **3** was new. Its spectral features showed the presence of one acetyl and two angeloyl residues. The relative position of the ester moieties was established with the demonstration that acetylation of the diester **5**, a compound of known constitution [7], gave a triester (**3**) identical with the natural product.

The high concentration of sesquiterpenoids in the roots of *L. garganicum* (*ca* 5% dry wt) meant that enough of lactones **3**–**6** were isolated for ^{13}C NMR determinations. Chemical shift and multiplicity considerations as well as comparison of the spectra of triesters **3** and **4** with those of diesters **5** and **6**, allowed assignment of all signals but that of C-13, at *ca* δ20. The resonance of this carbon and those of the methyl at C-2 of the angelate(s) and the methyl of



1



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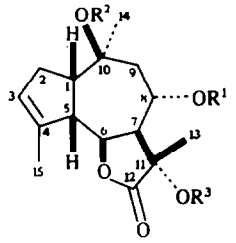
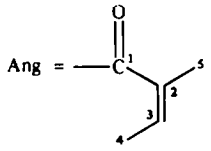
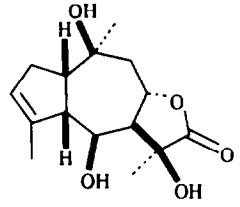


	R^1	R^2	R^3
3	Ang	Ac	Ang
4	Ang	Ac	Ac
5	Ang	H	Ang
6	Ang	H	Ac





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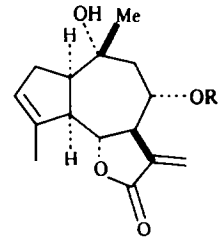

	R
8	Ac
9	H

the acetate(s) were in fact very close, within *ca* 0.8 ppm, and an unambiguous assignment was thus not possible. Apart from the substitutions at C-11 and C-13 (methyl hydroxy lactone *vs.* exomethylene lactone) and the ester moieties, the slovanolides **3–6** have the same constitution as cumabrins A and B (**8**, **9**) [8], but with a pseudoenantiomeric stereochemical arrangement at C-1, C-5, C-6 and C-10. Comparison of the ^{13}C NMR spectra of **3–6** and those of cumambrin derivatives [9], showed that the resonances of all these carbons are different. The major difference was at C-5 (*ca* 8 ppm), even though of the two couplings involving H-5, only $J_{1,5}$ was significantly different in compounds of the two series (5–6 Hz *vs.* 9–10 Hz).

The taxonomic relationship between *L. siler* and *L. garganicum* is not clear: Pignatti [2] recognizes *L. garganicum* as a species distinct from *L. siler* whereas Tutin [10] considers this plant as a subspecies of *L. siler*. The phytochemical pattern of *L. siler* and *L. garganicum* is similar, since compounds **2** and **4–6** co-occur in both plants, but differences are recognizable as regards the metabolism of phenylpropanoids and terpenoids, as neither laserin (**1**) nor the triester **3** were detected in *L. siler*. As regards both classes of compounds, a comparable infraspecific variability has, however, been found in *L. siler* [5], making it difficult to assess the taxonomic significance of the observed differences in chemical pattern between this plant and *L. garganicum*.

EXPERIMENTAL

Plant material. L. garganicum subsp. *garganicum* was collected near Monte Terminillo (Rieti) and identified by E.M.C. (voucher A1 at the Dipartimento Biologico, Università di Padova).

Isolation of compounds. Dried powdered roots (200 g) were extracted with $CHCl_3$ at room temp. A black syrup (29 g) was obtained, part of which (15 g) was chromatographed on a silica gel (50 g) column, eluted with petrol (50–70°) containing increasing amounts of $CHCl_3$; 50 ml fractions were collected. Fractions eluted with petrol–$CHCl_3$ (1:1) gave 900 mg of a mixture of **1**, **2** and **3**, 2.8 g of a mixture of **3** and **4**, 610 mg of **4** and 900 mg of a mixture of **5** and **6**.

All compounds were obtained in pure form by further chromatography on a silica gel column using mixtures of petrol–EtOAc (4:1 for the separation of **1–3**; 3:1 for the separation of **3** and **4**, and **5** and **6**). The yields were 39 mg **1**, 180 mg **2**, 860 mg **3**, 1.70 g **4**, 280 mg **5** and 190 mg **6**. Known compounds were identified by comparison of their physical and spectral data with those reported in the literature and, in the case of **5**, by comparison with an authentic sample.

8α,11α-Diangeloyloxy-10β-acetoxysloo-3-enolide (**3**). White amorphous powder, mp 68°, $[\alpha]_D^{25}$ −73.3° ($CHCl_3$; *c* 0.83); ^1H NMR (270 MHz, $CDCl_3$, TMS as reference): δ6.15 (*qq*, H-3'), 6.12 (*qq*, H-3"), 5.70 (*td*, *J* = 10.26, 10.26 and 0.6 Hz, H-8), 5.55 (*br s*, H-3), 4.77 (*dd*, *J* = 11.3 and 10.6 Hz, H-6), 3.75 (*t*, *J* = 10.6 Hz, H-7), 2.07 (*s*, OAc), *ca* 2.00 (*m*, H-4' + H-4"), *ca* 1.86 (*m*, H-5' + H-5"), 1.91 (*br s*, H-15), 1.62 (*s*, H-14), 1.53 (*s*, H-13); IR ν_{max}^{KBr} cm^{-1}:

Table 1. ^{13}C NMR data for compounds 3–6 (67.89 MHz, $CDCl_3$, TMS as int. standard)

C	3	4	5	6
1	49.04 *d*	49.17 *d*	49.21 *d*	49.01 *d*
2	31.42 *t*	31.71 *t*	31.99 *t*	31.92 *t*
3	125.43 *d*	125.55 *d*	125.61 *d*	125.64 *d*
4	145.80 *s*	146.03 *s*	146.89 *s*	146.69 *s*
5	47.68 *d*	47.89 *d*	47.28 *d*	47.16 *d*
6	78.02 *d*	77.60 *d*	78.07 *d*	78.04 *d*
7	53.37 *d*	53.42 *d*	55.72 *d*	55.67 *d*
8	63.56 *d*	64.14 *d*	66.62 *d*	66.59 *d*
9	40.62 *t*	40.97 *t*	43.47 *t*	43.25 *t*
10	82.54 *s*	82.64 *s*	71.08 *s*	71.00 *s*
11	77.83 *s*	78.23 *s*	78.15 *s*	78.04 *s*
12	173.25 *s*	173.83 *s*	174.11 *s*	173.66 *s*
13	20.06 *q**	20.16 *q**	20.08 *q**	20.70 *q**
14	24.51 *q*	24.70 *q*	30.85 *q*	30.75 *q*
15	18.26 *q*	18.14 *q*	18.61 *q*	18.54 *q*
Ang.				
1′	165.94 *s*	166.03 *s*	165.55/166.30 *s*	167.04 *s*
2′	127.15/126.47 *s*	127.30 *s*	126.96/126.57 *s*	126.66 *s*
3′	140.25/138.34 *d*	138.49 *d*	140.56/140.01 *d*	140.37 *d*
4′	15.60/15.44 *q*	15.60 *q*	15.77/15.72 *q*	15.68 *q*
5′	19.95/19.79 *q**	20.23 *q**	20.00/19.97 *q**	30.11 *q**
Ac.				
1″	169.59 *s*	170.25/169.51 *s*		169.15 *s*
2″	22.32 *q*	22.55/20.75 *q**		15.89 *q**

*Interchangeable signals.

1800 (α-acyloxy-γ-lactone), 1740 (acetate), 1725 (angelate); UV λ_{max}^{EtOH} nm (log ε): 215 (4.2); EIMS: no $[M]^+$ 428 $[M-HOAc]^+$ (2), 388 $[M-HOAng]^+$ (0.5), 328 $[M-HOAc-HOAng]^+$ (1), 228 $[M-HOAc-2HOAng]^+$ (30), 83 (100).

Saponification of 3–6 (*reaction of* 3 *is given as representative*). A 100 mg sample of 3 was solved in 5 ml of 5% methanolic KOH. After 16 hr the soln was diluted with H_2O (20 ml) and neutralized with 1% HCl. Extraction with $CHCl_3$ gave a yellowish oil (52 mg) which was purified through a short (5 g) column of silica gel eluted with petrol–EtOAc (1:3). Compound 7 (34 mg) was obtained, having physical and spectral data identical to those reported in ref. [6].

Acetylation of diester 5. A 131 mg sample of 5 was dissolved in 1 ml CH_2Cl_2 and 186 mg (5 mol equiv.) dimethylaminopyridine and 142 μl (5 mol equiv.) Ac_2O added. After 24 hr the reaction mixture was diluted with $CHCl_3$ (10 ml) and washed with dil. HCl, 5% $NaHCO_3$ and then dried. A yellowish oil (126 mg) was obtained. Purification by CC (10 g silica gel, petrol–EtOAc, 2:1) gave 59 mg triester 3, identical to the natural product.

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