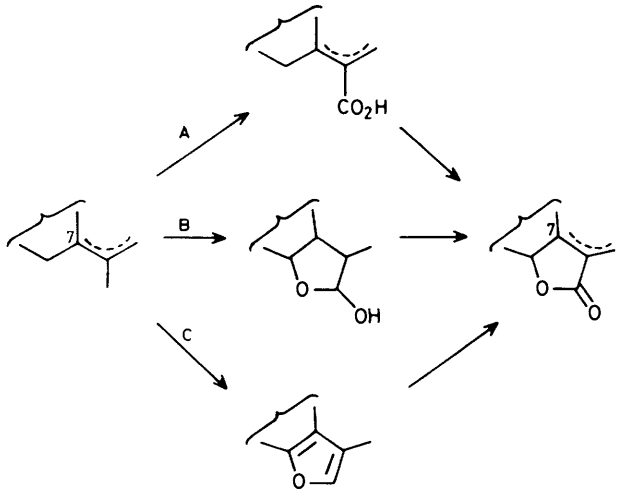
Structural and Conformational Studies on Sesquiterpenoid Esters from *Laserpitium halleri* Crantz subsp. *halleri*

Giovanni Appendino* and Maria Grazia Valle
Istituto di Chimica Farmaceutica e Tossicologica, Università di Torino, C. so Raffaello, 31, 10125 Torino, Italy
Pierluigi Gariboldi
Dipartimento di Chimica Organica ed Industriale, Università di Milano, Via Venezian, 21 20133 Milano, Italy

Investigation of the ripe fruits of *Laserpitium halleri* Crantz subsp. *halleri* afforded two pairs of new germacrane and guaiane esters, as well as the daucane derivative vaginatin, whose stereochemistry was assessed by spectral data and correlation with a compound of known configuration. The ^1H and ^{13}C n.m.r. spectra of the germacrane esters were characterized by the presence of sharp and broad signals. A conformational study of the natural compounds and several derivatives allowed us to relate the broadening of the n.m.r. lines to conformational motions which are discussed in detail. Among the derivatives of the natural esters, the ketone (**8**) was found to adopt, in solution, a conformation hitherto unreported for germacrane derivatives [C(4) and C(10) methyls *syn* on the α-face of the molecule, C(7) side-chain β-axial]. Comparison of the spectral features of some crossed/boat germacra-1(10),4-dienes and their corresponding 4,5-epoxides suggests that homoconjugation plays an important role in stabilizing the crossed/boat conformation of this class of cyclic 1,5-dienes. The biogenetic relationship between the compounds cumulated in the roots and the fruits of *L. halleri* is discussed.

The lactone group represents the most common oxidation level of the C(7)-isopropyl side-chain in cyclic *trans-trans* farnesylic sesquiterpenoids from most tribes of the *Compositae* family.[1] Intermediate states between the hydrocarbon and the lactone have occasionally been encountered, and the co-occurrence in some species of lactones and acids has led some to postulate that carboxylic acids are the ultimate precursors of lactones in these plant groups (Scheme 1, A).[2]



Scheme 1. Possible biogenetic routes to sesquiterpene γ-lactones

The *Laserpitieae* tribe of the *Umbelliferae* represents another major source of sesquiterpene lactones;[3] here, however, sesquiterpene carboxylic acids have never been reported. We recently isolated from the roots of the alpine plant *Laserpitium halleri* Crantz subsp. *halleri* the mixture of anomeric γ-lactols hallerin (**1**);[4] hallerin (**1**) is smoothly oxidized to lactone (**2**) and reduced to diol (**3**), both closely related to natural constituents of other *Laserpitieae*.† This suggests that γ-lactols might represent in this tribe an intermediate step in the oxidation of the side-chain to the lactone group (Scheme 1, B), with hydroxylation at C(6) or C(8) preceding at least the last oxidative step, as postulated for the furan route to eremophilanolides (Scheme 1, C).[5]

In this context, the isolation of further sesquiterpenoids from *L. halleri* seemed of interest. Investigation of the aerial parts showed that hallerin was present in all the plant except the ripe fruits; from the latter, five sesquiterpenoid esters were isolated.

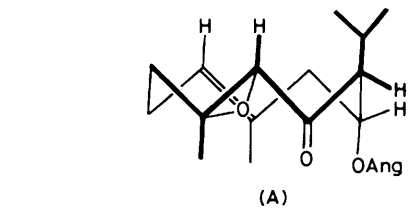
The least polar and most abundant compound was the diester (**4**). Its spectral features (Tables 1a and 2) were typical of a 4,5-epoxy-$\Delta^{1(10)}$-germacrane derivative bearing oxygenated functions at C(6) and C(8), and with the isopropyl side-chain *unoxidized*. The ester residues were identified as an acetate and an angelate‡ group by mass and n.m.r. spectroscopy. Their relative positions were established as follows: treatment of diester (**4**) with K_2CO_3 removed chemoselectively the acetate group, affording the monoester (**5**). Partial hydrolysis caused an upfield shift of the signal coupled with the oxirane proton, and oxidation of monoester (**5**) gave an epoxy ketone (**8**) in which the oxirane proton was a singlet. This located the acetate group at C(6).

In the ^1H n.m.r. spectrum of diester (**4**) some signals were rather broad at room temperature, particularly those of H(1), H(5), H(8), and H(9), and some *J*-values were unobtainable. Heating at 60 °C sharpened these signals, showing the presence of a conformational rate process. At neither temperature could stereochemical information be deduced from n.m.r. parameters.

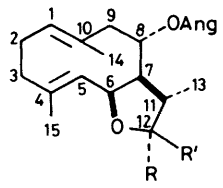
Esters of the parent germacranediol corresponding, exclusive of stereochemistry, to formula (**6**) have been reported from *Ferula ugamica*,[6] *F. rubroarenosa*[7] (family *Umbelliferae*), and from *Parabenzoin trilobum* Nakai[8] (family *Lauraceae*). Whereas the stereochemistry of esters from the genus *Ferula* is not known, the relative and absolute configuration of esters from *P. trilobum* has been established by a diffraction study,[9]

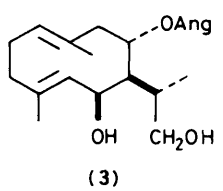
† The 11-angeloyloxy derivative of 8-deacyl lactone (**2**) has been isolated from *Laser trilobum* (L.) BORKH. (M. Holub, M. Budějšinský, Z. Smitalová, and D. Šaman, *Tetrahedron Lett.*, 1984, **25**, 3755); esters of 8-deacyl diol (**3**) occur in the genus *Thapsia* (G. Appendino, unpublished data).

‡ Angelate = (*Z*)-2-methylbut-2-enoate.



(A)

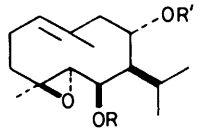


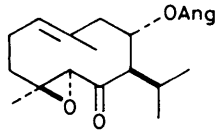


(3)

(1) R R: H OH; OH H

(2) =O





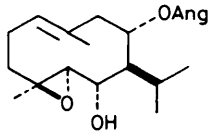
(8)

(4) Ac Ang

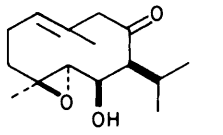
(5) H Ang

(6) H H

(7) Ac Ac



(9)



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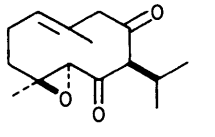

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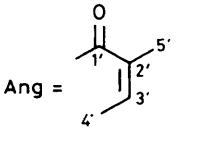


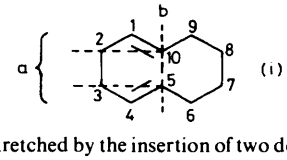
which revised a previous assignment.[10] Treatment of diester (**4**) with KOH gave a diol (**6**) which was acetylated to a diester (**7**) having physical and spectral data identical with those reported for a natural constituent of *P. trilobum*.[8] This established for the diester from *L. halleri* the relative and absolute configuration represented by formula (**4**).

A temperature-dependent line broadening comparable to that observed in compound (**4**) was also present in the ^1H and ^{13}C n.m.r. spectra of compounds (**5**)—(**7**), showing that all these compounds existed in solution at room temperature as a mixture of interconverting rotamers. The epoxy-ketone (**8**) was instead conformationally rigid, and displayed n.m.r. spectra consisting of sharp lines through the temperature range investigated (+25 to −50 °C). The proton–proton coupling constants showed that compound (**8**) adopted in solution a crossed/chair conformation* (A), with both methyls α and the side-chain β-axial. This represents a geometry which is thus far unreported for germacrane derivatives. Further evidence that (A) is the conformation in solution came from the stereochemical outcome of the reduction of compound (**8**). In conformation (A), the carbonyl group is roughly perpendicular to the mean plane through the carbocycle, and only one face of the carbonyl (*re*, leading to the 6α-alcohol) is available, the other being intra-annular and thus inaccessible to reagents (principle of peripheral attack[14]). In the other conformations possible for compound (**8**) (crossed/boat and parallel/half-chair), the carbonyl group is approximately parallel to the carbocycle; both sides are accessible, and no preferred attack is to be expected. Only one alcohol, (**9**), different from (**5**) and thus postulated as its epimer at C(6), was obtained when compound (**8**) was treated with $NaBH_4$, showing that (A) must be the conformation in solution. A diffraction study revealed that this is also the conformation in the solid state.[15]

The ^1H n.m.r. spectrum of compound (**9**) showed a broadening comparable to that of its epimer (**5**); in the ^{13}C n.m.r. spectrum, however, some signals were much broader, and clearly detectable only at 60 °C. ^{13}C T_1 Measurements showed values ranging from 0.5 to 4.0 s, thus excluding any involvement of this parameter in the abnormal line broadening observed (up to a half-weight width of *ca.* 50 Hz). A more marked and general broadening was present in the ^1H and ^{13}C n.m.r. spectra of the ketol (**10**) and the β-diketone (**11**), prepared from diol (**6**) by MnO_2[8] and pyridinium chlorochromate (PCC) oxidation respectively. The spectra could be interpreted in terms of structures (**10**) and (**11**) only at 60 °C, but even at this temperature some signals were still broad. The flexibility of compounds (**4**)—(**7**) and (**9**)—(**11**) led us to consider in a more general way the conformational aspects of the monoepoxides of germacra-1(10),4-dienes.

An epoxide ring is conformationally equivalent to a double bond, since it prevents rotation around the carbons bonded to the oxygen. The conformational analysis of the epoxides of germacradienes is, however, simpler than that of their dienic precursors.[11–13] Unlike a double bond, the oxirane ring cannot rotate through the ring, since this would place the oxygen inside the carbocycle, a situation impossible for medium sized rings. For the monoepoxides of germacradienes the interconversion operations[11,12] are thus limited to rotation of the endocyclic double bond through the ring, which inter-relates crossed and parallel orientations of the olefinic and the oxirane moieties, and to 'C(7)–C(8) inversions,'[11] a motion interconverting

* The *trans,trans*-1,5-cyclodecadiene system [(i), numbering as in germacrane derivatives] and its epoxides can be envisaged either as a



(i)

cyclohexane ring stretched by the insertion of two double bonds (a),[11] or as a decalin obtained by formal transannular cyclization through joining C(10) to C(5).[12,13] The different conformations of these systems can thus be described in two ways: one is based on the planar chirality of the double bonds and the geometry of the cyclohexane system obtained by notional migration of the C(2)–C(3) single bond between C(10) and C(5),[11] the other on the conformation of the cyclohexane rings of the decalin system.[12,13] The conformation of germacrane derivatives is given in this paper according to the former notation. Since in these compounds one double bond is epoxidized, thus having a real chirality, the relative orientation of the oxirane moiety and the double bond has been expressed by the descriptors 'parallel' (*anti*-methyls) and 'crossed' (*syn*-methyls). These terms are easy to visualize and independent of the substitution pattern of the carbons joined to the olefinic and oxirane moiety, even though they are strictly applicable only to conformations having a C(1)–C(2)–C(3)–C(4) torsion angle of 60° (crossed) and 0° (parallel). The possible conformations of 4β,5α-epoxy-$\Delta^{1(10)}$-*trans*-germacrane derivatives and their interrelationship are shown in Scheme 2.

Table 1. ^{1}H N.m.r. data[a]

(a)

	(4)		(5)		(6)	(7)		(8)	
		C_6D_6		C_6D_6			C_6D_6		C_6D_6
H(1)	5.32br t (8.0)	5.13br t (8.0)	5.32br t (8.0)	4.90br t (8.0)	5.14br t (9.0)	5.33br	5.05br t (9.0)	5.50br d (10)	4.98br dq (10; 1.0)
H(5)	2.86d (7.2)	2.96d (7.2)	2.82br d (8.0)	2.87br d (8.0)	2.76d (7.3)	2.85d (7.0)	2.91br d (7.0)	3.6s	3.27s
H(6)	4.90dd (7.2; 1.8)	5.30br d (7.2)	3.48dt (8.0; 8.0; 1.8)	3.62br d (8.0)	3.66dd (7.3; 1.5)	4.87d (7.3)	5.29dd (7.3; 1.5)	—	—
H(8)	5.49br dd (12; 5.0)	5.69br dd (12; 5.0)	5.20br dd (12; 5.0)	5.34qd (12; 5.0; 1.5)	4.22br dd (12; 4.0)	5.33br	5.59dd (12; 4.0)	5.60td (7.0; 7.0; 2.5)	5.60td (2.5; 7.0; 7.0)
H(9a)	2.60br	2.50br	2.60br	2.50br dd (14; 4.0)	2.55br	2.60br	2.65br	2.74br d(14)	2.44br dd (13.5; 2.5)
H(12)	0.89d (7.0)	1.02d (7.0)	0.94d (7.0)	0.93d (7.0)	0.92d (7.0)	0.90d (7.0)	1.02d (7.0)	1.04d (7.0)	0.82d (7.0)
H(13)	1.10d (7.0)	1.08d (7.0)	1.10d (7.0)	1.06d (7.0)	1.07d (7.0)	1.07d (7.0)	1.06d (7.0)	1.10d (7.0)	0.80d (7.0)
H(14)	1.76br s	1.46br s	1.78br s	1.42br s	1.71br s	1.73br s	1.44br s	1.69br s	1.53br d (1.0)
H(15)	1.14s	0.93s	1.14s	0.97s	1.11s	1.16s	0.93s	1.33s	1.34s
Others					3.40br (OH)			2.57dd (10; 7.0) [H(7)]	2.61dd (10; 7.0) [H(7)]

	(9)		(10)	(11)	(13)		(17)	(18)
		C_6D_6	C_6D_6[b]	C_6D_6[b]		C_6D_6[b]		
H(1)	5.25br m	5.02br dq (11.0; 1.5)	5.03br s	5.03br m	5.45br tq (8.0; 1.5)	5.09br tq (8.0; 1.5)		
H(5)	3.09d (4.0)	2.96d (4.0)	2.37d (7.0)	3.28s	2.30br (9.0)	2.77br d (9.0)		
H(6)	4.30dd (11.5; (4.0)	4.33dd (11.5; 4.0)	3.63m		4.23dd (9.0; 5.25)	4.01dd (9.0; 5.25)	5.08s	4.26dd (7.0; 2.1)
H(8)	5.25br m	5.33dq (12; 5.0; 1.0)	—	—	5.40br	5.42dq (11; 7.5; 3.5)	5.20dq (11.5; 4.5; 3.0)	5.20dq (7.5; 3.0; 4.5)
H(9a)		2.50br	2.83br d (12)	3.02d (12)	2.78br	2.60br d (12)		
H(12)	1.06d (6.5)	1.25d (6.5)	0.75d (6.5)	0.90d (6.5)	3.01dq (7.5; 3.0)	2.86dq (7.5; 3.0)	0.82d (7.0)	0.96d (7.0)
H(13)	1.10d (6.5)	1.25d (6.5)	0.84d (6.5)	0.99d (6.5)			0.90d (7.0)	1.08d (7.0)
H(14)	1.78br s	1.56br s	1.39br s	1.38br s	1.19d (6.5)	1.00d (6.5)	1.64br s	1.54br d (0.5)
H(15)	1.28s	1.27s	0.90s	0.90s	1.78br d (1.5)	1.36br d (1.5)	1.20s	1.18s
			3.60m (OH)	3.04d (6.0) [H(7)]	1.33s	0.92s		
			2.37br d (12) [H(9_β)]	2.64d (12) [H(9_β)]	3.01dq (7.5; 3.0) [H(11)]	2.86dq 7.5; 3.0) [H(11)]		

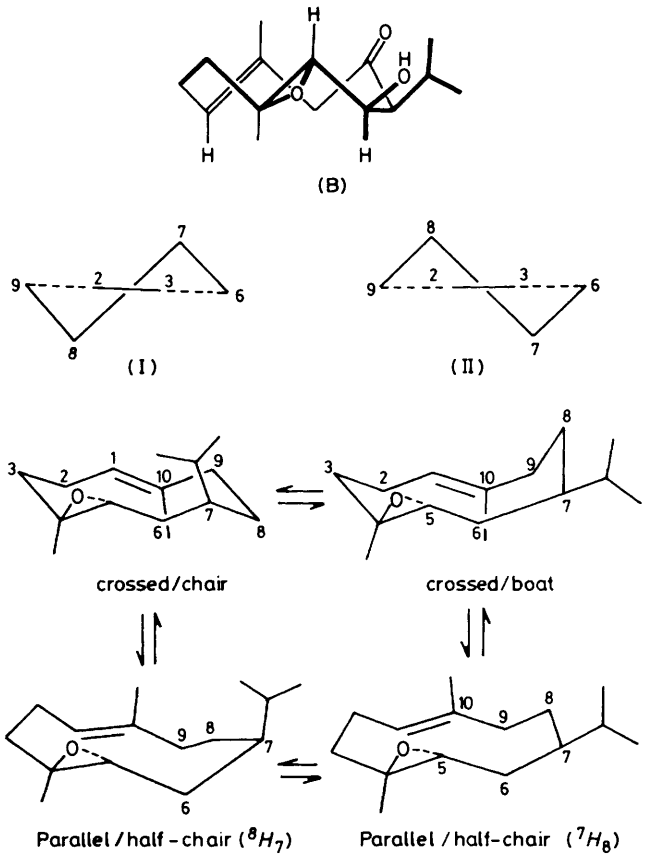
(b)

	(19)			(22)			(26)		(23)	
		C_6D_6	ΔTAI		C_6D_6	ΔTAI		ΔTAI		C_6D_6
H(1)	5.19d (7.8)	5.70d (7.8)	−0.16	5.47d (7.6)	5.74d (7.6)	−0.02	5.07d (6.7)	−0.12	4.28d (7.5)	4.18d (7.6)
H(2)	5.62br d (7.8)	5.83br d(7.8)	−0.11	5.53d (7.6)	5.66d (7.6)	−0.28	3.25d (6.7)	+0.06	5.55d (7.5)	5.50br d (7.6)
H(8)				4.02br	4.02dd (8.4; 6.0)	+1.68			3.94dd (5.7; 1.8)	3.71dd (6.0; 1.4)
H(12)	1.02d (7.2)	0.91d (7.2)	+0.16	1.00d (7.0)	1.02d (7.0)	−0.01	0.95d (7.2)	+0.11	0.97d (7.0)	0.98d (7.0)
H(13)	0.93d (7.2)	0.85d (7.2)	−0.07	0.93q (7.0)	0.98d (7.0)	−0.08	0.93d (7.2)	−0.04	0.91d (7.0)	0.95d (7.0)
H(14)	1.02s	1.05s	+0.21	1.04s	1.02s	+0.18	1.10s	+0.11	0.89s	0.87s
H(15)	1.70br s	1.46br s	−0.06	1.72br s	1.48br s	−0.07	1.30s	+0.03	1.72br s	1.56br d

[a] Spectra were taken at 200 MHz in $CDCl_3$ at 25 °C unless stated otherwise, with $SiMe_4$ as internal standard. Chemical shifts are given in δ-values, and J (in parentheses) in Hz. The proton signals for the angelate and the acetate group were almost identical for all the compounds. Those observed for compound (**4**) are taken as representative: (Ang): δ($CDCl_3$) 6.10 (qq, 7.0 and 1.5, 3′-H), 2.00 (dq, 7.0, 1.5, and 1.5, 4′-H_3), and 1.88(quint, 1.5 and 1.5, 5′-H_3). δ(C_6D_6) 5.78 (qq, 7.0 and 1.5, 3′-H), 1.98 (dq, 7.0, 1.5, and 1.5, 4′-H), and 1.86 (q, 1.5 and 1.5, 5′-H_3). Ac: δ($CDCl_3$) 2.03 (s). δ(C_6D_6) 1.70 (s). [b] At 60 °C.

geometries having C(7) up and C(8) down or C(7) down and (C(8) up relative to the mean plane through C(2), C(3), C(6), and C(9) (Scheme 2). For crossed conformations, C(7)–C(8) inversion flips the C(5)–C(10) fragment from a chair-like to a boat geometry (Scheme 2); for parallel arrangements, it interconverts two different half-chair geometries.* Independently from '(C(7)–C(8) inversion,' flipping from a crossed to a parallel conformation brings C(5), C(6), C(9), and C(10) on to

* These geometries are referred to in ref. 11 as twist; models show, however, that the conformations corresponding to a maximum staggering around the carbons C(6)–C(9) are better described, when C(10) is joined to C(5), as half-chair (two adjacent atoms out of the plane of the four others). The two half-chair geometries can be referred to as ($^8H_7$) and ($^7H_8$), meaning conformations with C(8) down and C(7) up or C(7) down and C(8) up relative to the plane of the other four atoms (J. C. P. Schwarz, *J. Chem. Soc., Chem. Commun.*, 1973, 505).



Scheme 2. Conformations of 4β,5α-epoxygermacra-1(10)-enes and their interconversion. (I) and (II) are views through an axis perpendicular to the C(2)–C(3) bond, illustrating 'C(7)–C(8) inversion' when crossed/chair and crossed/boat or parallel/half-chair ($^8H_7$) and parallel/half-chair ($^7H_8$) conformations are interconverted

the same plane, making the C(5)–C(10) fragment similar to a half-chair cyclohexane.

The ketol (**10**) adopts, in the solid state, an intramolecularly H-bonded parallel/half-chair ($^7H_8$) conformation (**B**).[16] In solution, rotamer equilibration takes place, at a rate intermediate, at room temperature, on the n.m.r. time-scale; in the ^{13}C n.m.r. spectrum some signals were in fact broadened beyond detection [C(10), C(14)], and others were only humps, slightly above the noise level [C(2), C(3), C(9)] (Figure 1). The i.r. spectrum showed two strong absorptions, at 1 680 and 1 660 cm^{-1}, corresponding to the intramolecularly H-bonded carbonyl of two distinct rotamers. At −60 °C some broad resonances in the ^1H n.m.r. spectrum separated into two unequal (*ca.* 3:1) sets of signals, showing that ketol (**10**) is birotameric in solution. Rotamer equilibration must involve rotation of the double bond through the ring, with the presence of H-bonded parallel/half-chair ($^7H_8$) and crossed/boat conformers. Other geometries of the C(5)—C(10) fragment do not allow the formation of H-bonding between the carbonyl group and the hydroxy group at C(6).

The β-diketone (**11**) showed spectral features similar to those of ketol (**10**); enolization as measured by n.m.r. spectroscopy was negligible both in $CDCl_3$ and in C_6D_6 (< 5%). From the extreme broadening, in the ^{13}C n.m.r. spectrum, of the signals of the carbons around the double bond, it is likely that this compound too exists in solution at room temperature as a mixture of crossed and parallel rotamers. However, the geometry of the fragment C(5)–C(10) could not be established.

For compounds (**4**)—(**7**) as well as their lactonized analogue (**13**), prepared from hallerin according to the method outlined in Scheme 3, line broadening was not so marked as in compounds (**10**) and (**11**), and was limited to certain signals. In the ^1H n.m.r. spectra of these compounds, the broadening pattern of the protons geminal to the three oxygen atoms was different, H(6) being much sharper than H(5) and H(8) (Figure 2, b). This situation must result from a molecular motion affecting the chemical shift of H(5) and H(8) to a greater extent than that of H(6), that is from the interconversion of rotamers having H(5) and H(8) in a different chemical environment. Models show that the observed line-broadening pattern is only consistent with the interconversion in solution of crossed/boat and parallel/half-chair ($^7H_8$) conformations, since they have H(5) subjected to different anisotropic shielding effects from the endocyclic double bond,[17] and H(8) orientated inside the ring. The crossed/chair and the parallel/half-chair ($^8H_7$) conformations have H(8) outside the ring, and presumably almost isochronous, whereas 'C(7)–C(8) inversion' brings H(6) into a 1,3-diaxial relationship with the oxygen function at C(8), and should hence mostly effect the chemical shift of this proton. Furthermore, in lactone (**13**), 'C(7)–C(8) inversion' is not expected to operate, owing to the reduced mobility of the C(5)—C(10) fragment, and yet the same broadening pattern observed in compounds (**4**)—(**7**) was present.

For compounds (**4**)—(**7**) the conformation in the solid state, which generally represents the most stable geometry, is different from that of ketol (**10**), even though in solution all these compounds exist as a mixture of crossed/boat and parallel/half-chair ($^7H_8$) rotamers. The monoester (**5**) crystallizes as a crossed/boat rotamer (C).[15] In spite of H-bonding between the hydroxy group and the carbonyl group of the angelate, the torsion angles round the carbocycle are very similar to those observed for the diester of compound (**6**) previously studied by *X*-ray methods [6-*O*-acetyl-8-*O*-(*p*-bromobenzoyl) derivative].[9] The crossed/boat conformation appears thus to be typical of compound (**6**) and its esters in the solid state.

Since the ketone (**8**) adopts the crossed-chair conformation, the oxidation of compound (**5**) must be accompanied by 'C(7)–C(8) inversion.' This represents the first case in which 'C(7)–C(8) inversion' has been shown to take place in germacrane derivatives with tetrahedral hybridization at C(7). The bulky isopropyl side-chain is expected to adopt a pseudo-equatorial orientation, and hence this operation, which interconverts pseudo-axial and pseudo-equatorial orientations of the substituents at C(7) and C(8), has generally been considered to be conformationally anchored in these compounds.[11]

It is interesting to note that, in the 1,5-dienes corresponding or analogous to the epoxides (**5**) and (**13**), the crossed/boat conformation is rigid, and these compounds [(**2**) and (**3**)] display n.m.r. spectra consisting of sharp lines unaffected by cooling (Figure 2, a). These differences appear to be related to homoconjugative interaction between the endocyclic double bonds.* In germacra-1(10),4-dienes, transannular overlapping of the double bonds is expected to stabilize the crossed and the parallel conformations, both having the planes of the double bonds in a parallel relationship, but not the transition state for their interconversion, in which these planes are perpendicular and no interaction is possible. Rotation of an endocyclic double bond through the ring should thus be more difficult in cyclodeca-1,5-dienes than in their monoepoxides, since the

* In cyclodeca-1,5-dienes, the delocalization energy associated with homoconjugation has been estimated as 5.5 kcal/mol (23 kJ/mol) by quantum mechanical calculations [J. Koutecky and J. Paldus, *Tetrahedron*, 1963, **19** (Suppl. 2), 201]. However, only parallel arrangements of the double bonds were considered.

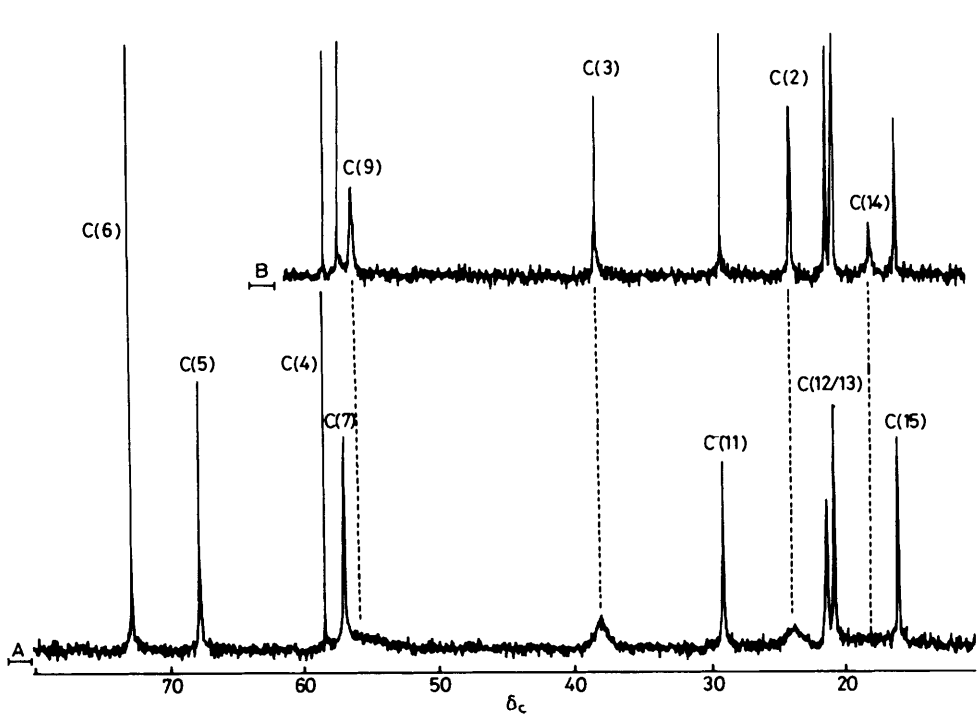
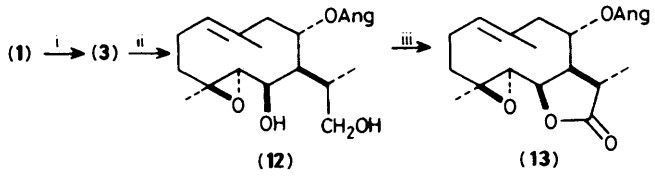


Figure 1. High-field part of the 50.30 MHz $^{13}C\{^1H\}$ spectrum of compound (**10**) (C_6D_6, $SiMe_4$ as internal standard) (A) 25 °C; (B) 60 °C



Scheme 3. Preparation of the epoxy lactone (**13**) from hallerin (**1**). *Reagents:* i, $NaBH_4$, MeOH; ii, TBHP, $VO(acac)_2$, CH_2Cl_2; iii, MnO_2. CH_2Cl_2

germacradienes are generally rigid (*cf.* costunolide- and parthenolide-type sesquiterpene lactones). In these epoxides the barrier for the rotation of the double bond through the ring must thus be higher, most probably on account of the geometry of the C(6)—C(10) fragment. When this fragment has a chair-like geometry, almost ideal staggering around its carbons exists; in the transition state for the interconversion of crossed and parallel conformations, eclipsing interactions are present along C(7)–C(8), and the transition state is thus destabilized. On the

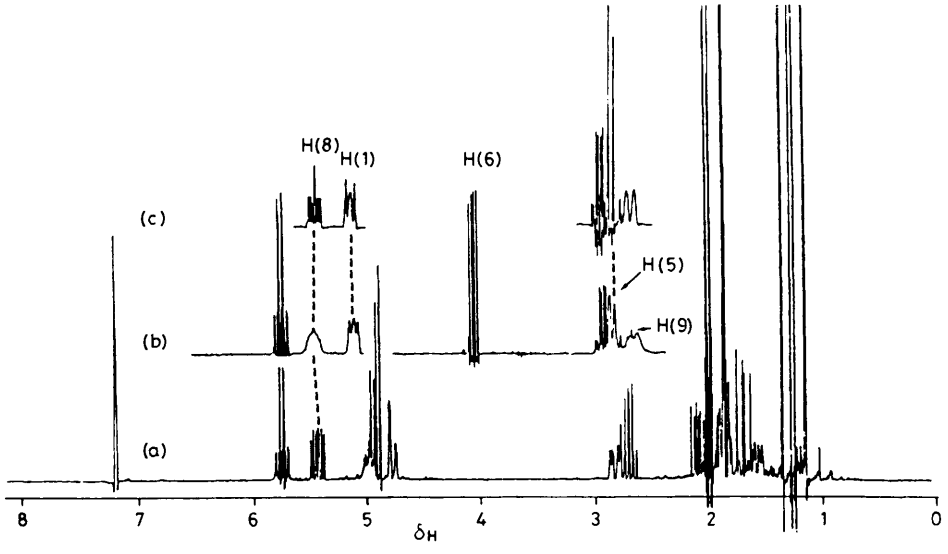


Figure 2. ^1H N.m.r. spectrum (C_6D_6) of (a) lactone (**2**) at 25 °C; (b) epoxy-lactone (**13**) at 25 °C; (c) epoxy-lactone (**13**) at 60 °C. Note that in (b) the signals of H(5) and H(8) are broad and that of H(6) is sharp

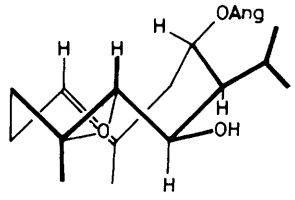
crossed and the parallel conformations are stabilized relative to the transition state for their interconversion.

In contrast with the flexibility of the 4,5-epoxides of crossed/boat germacradienes, the 4,5-epoxides of crossed/chair other hand, when the C(6)—C(10) fragment is in a boat-like geometry, an unfavourable eclipsing along C(7)–C(8) is already present. This makes the energy content of the C(6)—C(10) fragment more similar to that in the transition state, and thus

Table 2. ^{13}C N.m.r. data[a]

	(4)[ii]	(5)[ii]	(6)[ii]	(7)[ii]	(8)[ii]	(9)[i,§]	(10)[i,§]	(11)[i,§]	(13)[i]	(16)[i]	(17)[ii]	(19)[ii]	(22)[ii]	(23)[ii]	(27)[i,*]
C(1)	128.48d	128.77d	128.19d	128.44d	128.02d	128.30d	127.90d	125.00d	130.00d	127.64d	167.32s[y]	75.52d	72.80d	74.27d	74.93
C(2)	24.18t	24.29t	24.25t	24.16t	24.29t	24.84t	23.89t	25.00t	23.00t	23.50t	36.82t[†]	119.12d	122.30d	124.88d	59.68d
C(3)	38.22t	38.37t	38.55t	39.19t	39.76t[y]	38.60t	37.89t	36.40t	36.00t	37.40t	29.96t[x]	145.91s	143.23s	143.90s	62.83s
C(4)	59.14s	59.64s	60.42s	59.22s	64.82s	60.86s	59.18s	62.38s	60.42s	59.47s	79.56s	36.64t[‡]	35.30t	36.05t[‡]	30.84t
C(5)	66.46d	68.52d	69.31d	66.47d	64.44d	66.02d	67.70d	67.81d	61.76d	62.20d	46.77d	28.77t	29.62t	30.92t	27.32t
C(6)	73.74d	71.21d	71.93d	73.56d	204.85s	67.59d	72.34d	203.24s*	79.29d	77.38d	71.56d	81.78s	83.99s	84.94s	82.03s
C(7)	48.16d	50.58d	49.58d	47.84d	64.22d	48.08d	56.74d	65.08d	50.00d	43.99d	61.07d	59.89s	56.17s	58.13s	58.78s
C(8)	71.53d	72.14d	70.55d	71.83d	73.52d	73.28d	212.93s	199.50s*	70.00d	69.29d	69.50d	220.19s	80.04d	82.78d	219.56q
C(9)	41.62t	42.74t	44.65t	43.20t	36.62t[x]	41.31t	56.13t	54.00t	40.00t	43.06t	39.36t[†]	38.25t[‡]	37.12t	35.79t[‡]	37.93t
C(10)	130.87s	129.98s	132.00s	130.77s	132.15s	130.26s	130.62s	132.00s	130.00s	129.71s	125.26s[y]	50.58d	51.57d	55.48d	50.02s
C(11)	26.27d	26.16d	25.20d	26.19d	28.26d	26.98d	28.96d	28.73d	37.89d	134.41s	28.24d	25.96d	25.82d	2.731d	26.07d
C(12)	23.19q*	23.59q*	23.90q*	23.31q*	21.00q*,[y]	22.99q*	21.45q*	21.60q*	177.88s	168.16s	23.36q*	20.36q*	20.57q*	22.52q*	20.94q*
C(13)	21.31q*	21.37q*	21.45q*	21.19q*	19.63q*	19.24q*	20.92q*	19.35q*	15.76q	126.78t	21.26q*	25.79q*[†]	21.41q*	21.72q*	23.92q*
C(14)	20.38q	20.42q	20.55	20.34q	20.45q	20.67q	18.09q	18.80q	23.80q	19.45q	21.93q	17.97q	27.86q	26.15q	16.62q
C(15)	16.72q	16.32q	16.51q	16.77q	16.62q	15.95q	15.73q	15.28q	16.73q	15.82q	21.55q	25.29q*[†]	23.98q	24.92q	24.64q
Ang.															
C(1)′	166.52s	168.58s			166.51s	167.72s			166.38s		167.32s	165.76s	167.35s		165.70s
C(2)′	127.84s	127.15s			127.40s	127.72s			127.07s		128.00s	126.66s	127.51s		126.25s
C(3)′	137.85d	140.26d			138.94d	138.81d			139.85d		137.48d	138.51d	138.87d		139.60q
C(4)′	15.56q	15.90q			15.70q	15.77q			20.38q		20.58q	20.79q	20.57q		20.58q
C(5)′	20.68q	20.70q			20.74q	20.67q			15.73q		15.66q	15.35q	15.72q		15.70q
Ac															
C(1)″	169.56s			169.60s						168.16q	171.92s				
C(2)″	20.30q			20.74q, 15.70q						20.48q	21.46q				

[a] Spectra were taken at 25.18 (i) or 50.30 (ii) MHz, and at 25 °C in $CDCl_3$ with $SiMe_4$ as internal standard values unless stated otherwise. Chemical shifts are given in δ-values.*,†,‡; assignments with the same sign in the same column are interchangeable. Assignments bare based upon multiplicity and chemical-shift considerations, as well as inspection of the α-, β-, and γ-acylation shifts. Spectra marked with § were taken at 60 °C in C_6D_6; those marked with * in CD_3OD.



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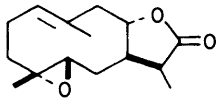

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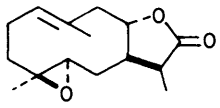

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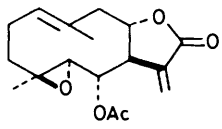


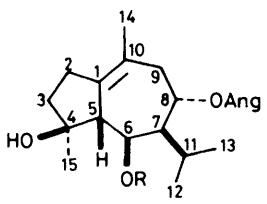
(16)

facilitates the interconversion of crossed and parallel conformations.*

These suggestions allow us to rationalize (in conformational terms) a certain number of observations reported in the literature. For instance, it has been reported that the 4α,5β-epoxides of *trans*-$\Delta^{1(10)}$-germacra-*trans*-(8)-olides (**14**) have sharp n.m.r. spectra, whereas their corresponding 4β,5α-diastereoisomers (**15**) display broad signals.[18] Only the 4α,5β-diastereoisomers can adopt the rigid crossed/chair conformation; for their 4β,5α-isomers, the corresponding crossed/chair conformation is impossible, since the isopropyl side-chain at C(7) and the oxygenated function at C(8) would be *trans*-diaxial, and the closure of the lactone ring would not take place [*cf.* (A)]. For these compounds conformational features similar to those of compounds (**4**)—(**7**) are therefore expected, with equilibration of crossed and parallel rotamers in solution. The 4β,5α-epoxy lactone spiciformin acetate (**16**) shows, in its ^1H and ^{13}C n.m.r. spectra, the same line-broadening pattern of compounds (**4**)—(**7**), and crystallizes in a crossed/boat conformation very close to that adopted by compound (**5**) in the solid state, (C).[16]

From more polar fractions of the extract of the fruits, the diester (**17**), the monoesters (**5**) and (**18**), and the daucane derivative (**19**) were also isolated. Compound (**5**) was identical with the monoester obtained from the partial saponification of diester (**4**); the compounds (**17**) and (**18**) were esters of a guaiane

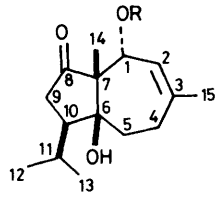
* Although germacrone 4,5-epoxide was shown by n.m.r. spectroscopy to adopt in solution a crossed/chair conformation, and no conformation rate process was evident (H. Hikino, C. Konno, T. Nagashima, T. Kohama, and T. Takemoto, *Tetrahedron Lett.*, 1971, 337), this compound gave pairs of isomeric rearranged products (caulolactones) and 1,10-epoxides upon treatment with $AlCl_3$ and peracids respectively (J. Endo, M. Nagasawa, H. Itokawa, and Y. Itaka, *Chem. Pharm. Bull.*, 1979, **27**, 275), thus requiring the equilibration of crossed and parallel rotamers, at least under the conditions of these reactions. The major flexibility of the crossed/chair conformation in germacrone 4,5-epoxide might be due to the presence of two adjacent sp^2 carbons in the fragment C(6)—C(10).



R

(17) Ac

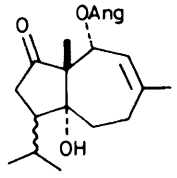
(18) H



R

(19) Ang

(20) Ac



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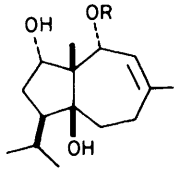
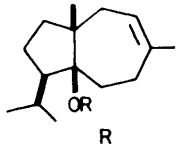


R

(22) Ang

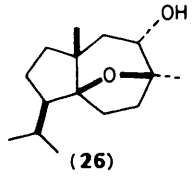
(23) H



R

(24) H

(25) Ac



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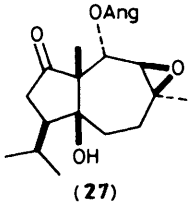

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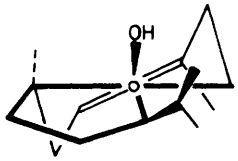

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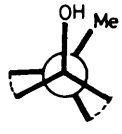
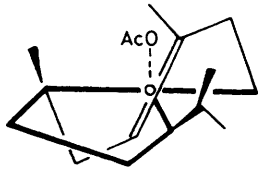
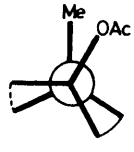




(E)



triol. Spectral data, as well as the results of the partial saponification, established that both these compounds had an angeloyl residue at C(8), whereas a C(6) compound (**17**) had an acetyl group, and compound (**18**) an hydroxy group. The relative and absolute configuration was established by correlation with compound (**4**). Treatment of this germacrane diester with BF_3-Et_2O gave, in 30% yield, a guaiane diester identical with the natural compound (**17**).

The daucane derivative was the ketol vaginatin, previously isolated from the roots of *Selinum vaginatum* C. B. Clarke.[19] To this compound was assigned formula (**19**), exclusive of stereochemistry.[20] The ketol from *L. halleri*, a crystalline solid, had similar $[\alpha]_D$ and spectral features (^1H and ^{13}C n.m.r., m.s.) to those reported for a daucane derivative isolated as an oil from *Inula crithmoides* L. (family *Compositae*),[21] and to which stereostructure (**21**) was assigned. However, the *trans*-junction of the bicyclic system was not consistent with the acylation shifts observed upon '*in situ*' carbamoylation of compound (**19**) with trichloroacetyl isocyanate (TAI).[22] The large downfield shift of the angular methyl and one methyl of the isopropyl side-chain (Table 1b) required a *syn* orientation of these groups with the tertiary hydroxy group, and therefore a *cis* junction of the hydroazulenic system. This was further confirmed by the conversion of compound (**19**) into a triol (**23**), identical with a compound prepared from siol acetate (**20**), a derivative of known relative configuration at C(1), C(6), C(7), and C(10).[23]*

The *cis*-hydroazulenic system of daucanes such as compounds (**19**) and (**20**) can exist in two main conformations, typified by those of carotol (**24**) and carotol acetate (**25**). In carotol the 7-membered ring adopts the twist-chair conformation (D), with the hydroxy group H-bonded with the double bond;[24] in carotol acetate this ring is instead in the twist-boat conformations (E).[24] These two geometries can be distinguished *via* the outcome of the epoxidation of the double bond:[24,25] compounds adopting the carotol-type conformation give daucol-type hydroxy ethers (**26**) as a result of transannular opening of an intermediate epoxide from the tertiary hydroxy group, whereas compounds adopting the carotol acetate-type conformation give epoxy derivatives, since the hydroxy group and the epoxide are not properly orientated to interact.[25]

Treatment of compound (**19**) with *meta*-chloroperbenzoic acid (MCPBA) gave the epoxy derivative (**27**), establishing that the conformation of compound (**19**) in solution is of the carotol acetate-type.† The formation of only one isomer can be ascribed to the presence of an oxygen function allylic to the double bond. In cyclic olefins, allylic ester groups orientate the epoxidation '*anti*,'[26] and thus the epoxide ring in the product (**27**) was placed β to the ring system.

The reduction of compound (**19**) with $NaBH_4$ was completely stereoselective, affording only one of the two possible diastereoisomers at C(8). The *cis*-hydroazulenic system is a 'bowl-shaped' molecule, and approach of reagents to groups located on carbons near the ring junction is expected to occur mostly from outside the bowl, *i.e.* from the same side as the ring-junction substituents (β-face). For compound (**19**), approach from the internal side (face α) is further hindered by the angeloyl residue at C(1). The newly formed hydroxy group was thus expected to be the result of the attack from the external (β) face, and was formulated as having the α configuration at C(1).

Treatment of angelate (**22**) with $LiAlH_4$ afforded the triol (**23**). Removal of the angeloyl residue markedly affected the

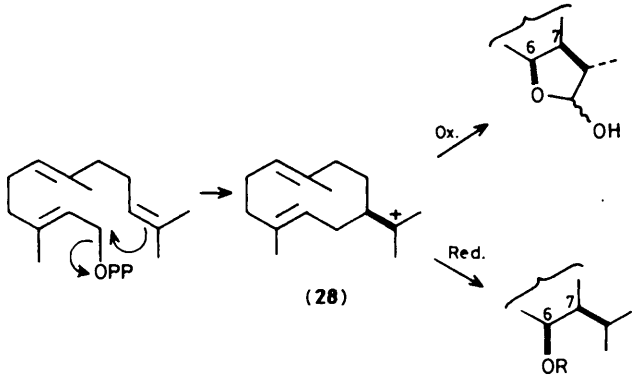
* The daucane skeleton does have a universally accepted trivial numbering, and several different systems can be found in the literature, none of them complying with the systematic rules for the azulene skeleton. We adopted the numbering used for the daucane esters from *Ferula linkii* Webb (J. G. Diaz, B. M. Fraga, A. G. Gonzalez, P. Gonzalez, and M. G. Hernandez, *Phytochemistry*, 1984, **23**, 254), since the sequence of carbons around the carbocycle follows a biogenetic order.

† The steric outcome of a reaction can be related to the conformation of a reagent only if no conformational rate process takes place in the latter, so that the Curtin–Hammett principle does not operate. According to its spectral features, compound (**19**) is monorotameric in solution [*cf.* the reduction of ketone (**8**)].

multiplicity pattern of some signals, particularly that of H(8) (Table 1a), showing that the conformation had changed, most probably because of differences in H-bonding pattern. In the carotol acetate-type conformation, α-orientated oxygen atoms at C(1) and C(8) are not properly disposed for the formation of an H-bond of 6-membered geometry. In ester (**24**), the H-bond takes place presumably through an 8-membered geometry, with the carbonyl group of the acyl residue acting as the acceptor, as observed in the solid state for compound (**5**).[15] In triol (**23**) however, the 6-membered geometry is the only one possible; flipping to the carotol-type conformation, which can accommodate a bond of this type, thus takes place. As a result, the two hydroxy groups approach, and formation of a second H-bond is also possible between the tertiary hydroxy group and the double bond, whose acceptor capacity is increased by removal of the allylic electron-withdrawing acyl group.

The absolute configuration of compound (**19**) could not be established; a positive Cotton effect (C.e.) for the n—π^* transition of the carbonyl group shed no light on the absolute configuration, since it was difficult to assess the relative influence of the substituents on the sign of the C.e.

From the biogenetic point of view, compound (**19**) is not directly related to hallerin (**1**), since the daucane skeleton is of *cis,trans*-farnesyl derivation. The germacranes (**4**) and (**5**), the guaianes (**17**) and (**18**), share instead (with hallerin) a *trans,trans*-farnesylic origin. The cyclization of *trans,trans*-farnesyl pyrophosphate to a cyclic germacrane intermediate (**28**) involves the formation of a positive charge on the side-chain (Scheme 4). Further modification of this moiety can take



Scheme 4. Metabolism of the C(7)-isopropyl side-chain in *Laserpitium halleri*

place in *L. halleri* in two ways: an oxidative path leads to hallerin, which has the side-chain lactolized, whereas a reductive path leads to esters (**4**), (**5**), (**17**), and (**18**), all having the side-chain reduced to an isopropyl group.

The exact site of synthesis of terpenoids in plants is often not known;[27] it is, however, worth nothing that, in *L. halleri*, compounds derived from distinct metabolic pathways, at least as regards the isopropyl side-chain, are accumulated in different plant parts: hallerin in the roots, and compounds (**4**), (**5**), (**17**), and (**18**) in the fruits.

Experimental

For general instrumentation, see ref. 4.

Plant Material.—Fruits of *L. halleri* were collected near Lillaz (Cogne, Valle d'Aosta, Italy) during late September in the years 1981—1983. Plant material was identified by P. A. S. Stefenelli and Dr. L. Poggio.

Isolation of the Constituents.—(*From the* 1982 *collection*). Dried, powdered fruits (720 g) were extracted with $CHCl_3$ at room temperature. Removal of the solvent left an oil (36 g), part of which (20 g) was chromatographed on a silica gel (1 kg) column, which was eluted with mixtures of light petroleum (b.p. 50—70 °C) and $CHCl_3$. The fractions eluted with a 1:9 mixture gave compound (**4**) (900 mg), (**19**) (300 mg), (**17**), (16 mg), (**5**) (200 mg), and (**18**) (30 mg).

Shiromodiol-6-O-acetate-8-O-angelate (**4**). This was obtained as an *oil*, $[\alpha]_D^{25}$ −54° ($CHCl_3$, *c* 1.1); $\nu_{max.}$($CHCl_3$) (no OH band), 1 750—1 250 (acetate), and 1 720 cm^{-1} (unsaturated ester): $\lambda_{max.}$ 218 nm (log ε 3.9); *m/z* (rel int.) M^+, 378.240 594 (2%) ($C_{22}H_{34}O_5$ requires *M*, 378.246 09) and 83 ($C_5H_7O^+$, 100).

Shiromodiol-8-O-angelate (**5**). *Crystals* (from light petroleum–$CHCl_3$), m.p. 113—114 °C; $[\alpha]_D^{25}$ −57° ($CHCl_3$, *c* 1.04); $\nu_{max.}$(KBr) 3 420 (OH) and 1 720 cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.8); *m/z* (re. int.) M^+, 336.230 200 (1%) ($C_{20}H_{32}O_4$ requires *M*, 336.230 045) and 83 (100).

Cycloshiromodiol-6-O-acetate-8-O-angelate (**17**). An *oil*, $[\alpha]_D^{25}$ +112° ($CHCl_3$, *c* 0.90); $\nu_{max.}$($CHCl_3$) (no OH band), 1 740 and 1 245 (acetate), and 1 705 cm^{-1}; $\lambda_{max.}$ 214 nm (log ε 3.9); *m/z* (rel. int) M^+ 378.240 601 (1%) ($C_{22}H_{34}O_5$ requires *M*, 378.240 609) and 83 (100).

Cycloshiromodiol-8-O-angelate (**18**). *Crystals*, m.p. 118—119 °C; $[\alpha]_D^{25}$ +96° ($CHCl_3$, *c* 0.90); $\nu_{max.}$(KBr) 3 400 and 1 715 cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.9); *m/z* (rel. int.) M^+, 336.230 180 (<1%) ($C_{20}H_{32}O_4$ requires *M*, 336.230 045) and 83 (100).

Vaginatin (**19**). Crystals, m.p. 68—69 °C (lit.,[19] 77—78 °C;[21] oil); $[\alpha]_D^{25}$ −247° ($CHCl_3$, *c* 1.5) (lit.,[19] −266.7°; −212.8°); $\nu_{max.}$(Nujol) 3 420 (OH) and 1 720 cm^{-1} (unsaturated ester); $\lambda_{max.}$ 218 nm (log ε 3.9); *m/z* (rel int.) M^+, 334.214 298 (1%) (Calc. for $C_{20}H_{30}O_4$: *M*, 334.214 396) 83 (100); c.d. (MeOH) $\Delta\varepsilon_{305}$ −0.11; $\Delta\varepsilon_{215}$ −23.38.

Mild Saponification of Compound (**4**).—A sample of compound (**4**) (151 mg) was dissolved in 50% aqueous methanol (5 ml), and the resulting solution was saturated with K_2CO_3. After the mixture had been stirred for 24 h at room temperature, water (20 ml) was added and the reaction mixture was extracted with $CHCl_3$. The extract was washed with water and dried ($MgSO_4$). Evaporation of the solvent gave an oil (102 mg) which was chromatographed on a short column of silica gel (10 g) and eluted with light petroleum–ethyl acetate (4:1). Monoester (**5**) (62 mg) was obtained, whose m.p., $[\alpha]_D^{25}$, and spectral properties were identical with those of the natural ester (**5**) isolated previously.

Oxidation of Monoester (**5**).—Compound (**5**) (96 mg, 0.28 mmol) was dissolved in dry CH_2Cl_2 (5 ml) and PCC (119 mg, 0.56 mmol) was added. After being stirred for 3 days at room temperature, the reaction mixture was diluted with CH_2Cl_2 (20 ml) and filtered through a short pad of silica gel: a crystalline material (75 mg) was obtained. Recrystallization from hexane gave the ketone (**8**) (58 mg) as needles, m.p. 109—110 °C; $[\alpha]_D^{25}$ +15.9° ($CHCl_3$, *c* 2.0); $\nu_{max.}$ (no OH bands), 1 705 cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.9); *m/z* (rel. int.) M^+ 334.213 867 (1%) (Calc. for $C_{20}H_{30}O_4$: *M*, 334.214 396) and 83 (100).

Saponification of Diester (**4**) *to Diol* (**6**).—A sample of diester (**4**) (650 mg) was dissolved in 5% methanolic KOH (10 ml); after 24 h, water (50 ml) was added, and the solution was extracted with CH_2Cl_2 to give an oil (320 mg). The oil was purified by chromatography on a silica gel column (20 g) and eluted with

• The $[\alpha]_D$ value of diol (**6**) is not given in the papers on its esters (ref. 8 and 10); in a collection of physical data on terpendoids, diol (**6**) is erroneously reported as being laevorotatory {$[\alpha]_D^{25}$ −33.3° ($CHCl_3$) (J. S. Glasby in 'The Encylopaedia of the Terpenoids,' Wiley, 1982, p. 2198)}.

$CHCl_3$–ethyl acetate (6:1); the diol (**6**) (220 mg) was obtained, m.p. 86—87 °C (lit.,[8] 89 °C); $[\alpha]_D^{25}$ +53.8° ($CHCl_3$, *c* 1.3).*

Acetylation of Shiromodiol (**6**).—A shiromodiol (**6**) (50 mg) was treated overnight with a mixture of pyridine and Ac_2O (1 ml). The reaction mixture was diluted with water (10 ml), and extracted with CH_2Cl_2. The organic phase was washed successively with 5% aqueous $NaHCO_3$, dilute HCl, and water, and then dried ($MgSO_4$) to afford, after work-up, the diacetate (**7**) (52 mg). Crystallization from CH_2Cl_2–hexane gave needles, m.p. 110—111 °C (lit.,[8] 109—112 °C); $[\alpha]_D^{25}$ −55.8° ($CHCl_3$, *c* 1.01((lit.,[8] −61.9.°).

Reduction of Ketone (**8**).—A ketone of (**8**) (80 mg) was dissolved in methanol (5 ml) and the solution was treated with $NaBH_4$ (20 mg). After being stirred at room temperature for 5 min, the reaction mixture was diluted with water (20 ml), neutralized with HOAc, and extracted with CH_2Cl_2. The organic phase was washed with 5% aqueous $NaHCO_3$ and dried ($MgSO_4$). Removal of the solvent gave an oil (72 g) which was purified by chromatography on a silica gel (5 mg) column; the hydroxy ester (**9**) (63 mg) was obtained as an oil, $[\alpha]_D^{25}$ −58.3 ($CHCl_3$, *c* 1.8); $\nu_{max.}$($CHCl_3$) 3 400 and 1 720 cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.9); *m/z* (rel. int.) M^+ 336.231 195 (< 1%) ($C_{20}H_{32}O_4$ requires 336.230 045) and 83 (100).

Oxidation of Shiromodiol (**6**) *with Active* MnO_2.—A sample of shiromodiol (**6**) (102 mg) was dissolved in CH_2Cl_2 (10 ml) and the solution was stirred at room temperature with active MnO_2 (500 mg). After 1 h the mixture was centrifuged and the deposit of MnO_2 was washed twice with acetone. The pooled organic phases were evaporated to give the hydroxy ketone (**10**) (90 mg). Crystallization from hexane gave needles (77 mg), m.p. 94—95 °C (lit.,[8] 95—96 °C); $[\alpha]_D^{25}$ −273° ($CHCl_3$, *c* 1.6); $\nu_{max.}$(KBr) 1 680 and 1 660 cm^{-1}; *m/z* (rel. int.) M^+, 252.172 551 (2%) (Calc. for $C_{15}H_{24}O_3$: 252.172 534).

Acetylation in the usual manner gave a crystalline monoacetate, m.p. 100—101 °C; $[\alpha]_D^{25}$ −288° ($CHCl_3$, *c* 1.9), in which the i.r. absorption of the keto group was shifted to the normal position (1 705 cm^{-1}, broad band both in KBr disc and in $CHCl_3$ solution).

β-*Diketone* (**11**).—(a) *From shiromodiol* (**6**). A solution of shiromodiol (**6**) (100 mg) in dry CH_2Cl_2 (2 ml) was treated with an excess of PCC (200 mg); after 4 d the reaction mixture was worked up as described for the oxidation of compound (**5**), to give the dione (**11**) (35 mg) as needles, m.p. 75 °C (lit.,[8] 84—85 °C); $[\alpha]_D^{25}$ −257° ($CHCl_3$, *c* 1.6); *m/z* (rel. int.) M^+, 250.156 906 (1%) (Calc. for $C_{15}H_{22}O_3$: *M*, 250.156 885).

(b) *From ketol* (**10**). A solution of compound (**10**) (79 mg) in dry CH_2Cl_2 (3 ml) was treated with PCC (100 mg). After 4 d the reaction mixture was worked up as described for the oxidation of compound (**5**), to give dione (**11**) (50 mg).

Lactone (**13**) *from Hallerin* (**1**).—(i) To a magnetically stirred solution of hallerin (**1**) (1.388 g) in methanol (20 ml) was added $NaBH_4$ (620 mg). After 5 min the solution was worked up as described for the reduction of compound (**8**), to give a yellowish oil (1.013 g). Purification by column chromatography on a silica gel (25 g) column eluted with light petroleum (50—70)–ethyl acetate (4:1) gave diol (**3**) (660 mg) as an oil, $[\alpha]_D^{25}$ −92° ($CHCl_3$, *c* 0.55). (ii) A solution of diol (**3**) (340 mg, 1.01 mmol) in CH_2Cl_2 (6 ml) was treated with 70% aqueous butyl hydroperoxide (TBH) (251 μl, 2 mmol) and $VO(acac)_2$ (15 mg) at room temperature. After 4 h the reaction mixture was diluted with CH_2Cl_2 (30 ml) and washed successively with 10% aqueous Na_2SO_3 and brine. Evaporation of the solvent gave a yellowish oil (336 mg), which was purified by column chromatography on silica gel (10 g). Elution with light petroleum (50—70)–ethyl acetate (1:1) afforded the epoxide (**12**) (286 mg) as a white powder, m.p. 111 °C; $[\alpha]_D^{25}$ −42° (acetone, *c* 1.1). Analogous results were obtained when TBHP was employed anhydrous or as an 80% solution in t-butyl peroxide.

(iii) A sample of the epoxy diol (**12**) (280 mg) was dissolved in CH_2Cl_2 (10 ml), and active MnO_2 (3 g) was added; after 6 h the solution was centrifuged and the residue of MnO_2 was washed twice with acetone. Evaporation of the pooled organic phases gave the lactone (**13**) (208 mg). Crystallization from hexane–$CHCl_3$ gave needles, m.p. 129—130 °C; $[\alpha]_D^{25}$ −51° ($CHCl_3$, *c* 0.80); $\nu_{max.}$ (no OH band), 1 785 (γ-lactone), and 1 720 (unsaturated ester) cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.6); *m/z* (rel. int.) M^+ 348.194 962 (3%) (Calc. for $C_{20}H_{28}O_5$: *M*, 348.193 661) and 83 (100).

Transannular Cyclization of Compound (**4**).—A solution of compound (**4**) (420 mg) in dry ether (10 ml) was cooled at 0 °C and treated with freshly distilled $BF_3{\cdot}Et_2O$ (1 ml). After 2 h the solution, which had turned deep red, was poured into 5% aqueous $NaHCO_3$ (20 ml), and extracted with ethyl acetate. The extract was washed with brine and dried. Removal of the solvent gave a gummy residue (260 mg), which was purified by column chromatography on silica gel (10 g). Elution with light petroleum (50—70)–ethyl acetate (3:1) gave the diester (**17**) (128 mg), identical ($[\alpha]_D^{25}$, i.r., ^1H n.m.r.) with the natural product.

Mild Saponification of Diester (**17**).—A solution of compound (**17**) (50 mg) in 50% aqueous methanol (5 ml) was treated with K_2CO_3 as described for the mild saponification of compound (**4**), to give a monoester (23 mg), identical ($[\alpha]_D^{25}$, m.p., i.r.) with natural compound (**18**).

Epoxidation of Vaginatin (**19**).—A solution of vaginatin (**19**) (286 mg, 0.85 mmol) in CH_2Cl_2 (5 ml) was treated with MCPBA (85%; 209 mg, 1.0 mmol). After being stirred for 7 days at room temperature, the solution was diluted with CH_2Cl_2, washed successively with 5% aqueous $NaHCO_3$ and water, and dried ($MgSO_4$). Removal of the solvent gave an oil (266 mg) which was purified through a short column of silica gel (10 g). Elution with light petroleum (50—70)–ethyl acetate (2:1) afforded unchanged vaginatin (**19**) (34 mg) and the epoxide (**27**) (160 mg) as an oil, $[\alpha]_D^{25}$ −68.4° ($CHCl_3$, *c* 1.8); $\nu_{max.}$ 3 400 and 1 710 cm^{-1}; $\lambda_{max.}$ 218 nm (log ε 3.8); *m/z* (rel. int.) M^+ 350.214 023 ($C_{20}H_{30}O_5$ requires 350.209 310) (1%) and 83 (100).

Reduction of Vaginatin (**19**).—A solution of vaginatin (**19**) (79 mg) in methanol (5 ml) was treated with $NaBH_4$ (30 mg). After 5 min, water was added (20 ml), followed by dilute HCl (2 drops), and the solution was extracted with CH_2Cl_2. Evaporation of the extract afforded an oil (68 mg) which was purified by preparative t.l.c. [eluant $CHCl_3$–acetone (6:1)] to give compound (**22**) (56 mg) as an oil, $[\alpha]_D^{25}$ −140° ($CHCl_3$, *c* 1.6); $\nu_{max.}$(Nujol) 3 500 and 1 710 cm^{-1}; *m/z* (rel. int.) 336 (M^+, < 1%) and 83 (100).

Reductive Cleavage of Ester (**22**).—A solution of compound (**22**) in dry ether (7 ml) was treated with an excess of $LiAlH_4$ (120 mg). After being stirred for 45 min at room temperature, the reaction mixture was worked up by the addition of a few drops of ethyl acetate followed by saturated aqueous $MgSO_4$ (30 ml). Extraction of the mixture with ethyl acetate gave a solid material (255 mg), which crystallized from di-isopropyl ether–light petroleum (50—70) to give triol (**23**) as needles, m.p. 123—125 °C (lit.,[19] 122 °C;[23] 123—125 °C); $[\alpha]_D^{25}$ −65.3° (acetone, *c* 1.16) (lit.,[19] −80.6°C[23] −57 °C); $\nu_{max.}$(KBr) 3 400 cm^{-1}, no carbonyl absorption; *m/z* (rel. int.) 254 (M^+, < 1%) and 139 (100).

Acknowledgements

This work was supported in part by the Ministero della Pubblica Istruzione (Fondi 60 and 40%) and by the Assessorato Agricoltura e Foreste, Servizi Forestali della Regione Autonoma Valle d'Aosta. We are grateful to Prof. G. M. Nano for his encouragement and useful advice. We thank Drs. C. Bicchi and C. Frattini for the mass spectra, and Dr. L. Poggio (Giardino Botanico Alpino Paradisia, Valnontey, Cogne) for the plant material.

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Received 5th August 1985; *Paper* 5/1344