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NEW SESQUITERPENE LACTONES AND ACETYLENES FROM *CHRYSANTHEMUM LAVANDULIFOLIUM*

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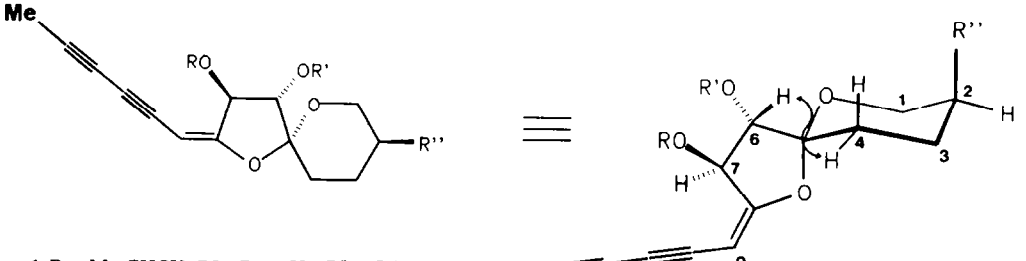
Abstract- The aerial parts of *Chrysanthemum lavandulifolium* Mak. (Asteraceae) yielded three new acetylenes of the spiroacetal type **1-3**, as well as two new germacranolides **4-5** and several known lactones.

The genus *Chrysanthemum*, of the family Asteraceae (Compositae), tribe Anthemideae, has not been fully delimited from the botanical point of view. Definitive adscriptions of many species either to the genus *Chrysanthemum* or to related genera such as *Tanacetum*, *Leucanthemum*, etc. remain controversial; in *Flora Europaea*[1] only three species are categorized under *Chrysanthemum*. Polyacetylenic compounds are the most characteristic products of this genus;[2] other typical Compositae metabolites such as sesquiterpene lactones[3] or coumarins[4] are found much less frequently. The present paper reports on the results of our investigation of *Chrysanthemum lavandulifolium* Mak., a species which grows in Eastern Asia. Extraction of aerial parts of the plant and subsequent chromatographic fractionation of the extract yielded five new natural metabolites, together with several known ones. The structures of the new compounds have been established, as described below, with the aid of spectroscopic techniques, mainly that of high-field NMR spectroscopy.

Compound **1** has the molecular formula $C_{21}H_{26}O_7$. The IR spectrum of the compound shows bands characteristic of OH (3450 cm^{-1}), C≡C bonds (2229, 2139 cm^{-1}) and ester groups (1733 cm^{-1}). Furthermore, the pattern of the UV spectrum (see data in the Experimental part) suggests the bonding of a conjugated ene-diyne unit to a cyclic spiroacetal, a structural feature[2] of numerous natural acetylenes from Anthemideae.

The high-field ^1H and ^{13}C NMR spectra (Tables 1 and 2) provide more insight into the molecular structure of the compound, as they resemble those of known spiroacetal enol ethers.[2,5-8] The three-proton doublet (J = 1 Hz) at δ 1.93 in the ^1H NMR spectrum can be assigned to the terminal methyl group of an ene-diyne moiety (see below). Irradiation of this doublet caused the collapse of a complex signal at δ 5.12 into a doublet with J = 2 Hz. This signal was thus assigned to the olefinic hydrogen of the enol ether fragment. As was to be expected, irradiation of this signal transformed the methyl doublet into a singlet and also simplified a double doublet at δ 5.99 into a doublet with J = 7 Hz. This signal is further coupled to a doublet (J = 7 Hz) at δ 3.89, as was deduced from spin decoupling. All these facts confirm the existence of the structural fragment CH_3-C≡C-C≡C-CH=C(OR)-CH-CH-C. As

regards the ^{13}C NMR spectrum (Table 2), it displays twenty-one signals, in agreement with the molecular formula. The multiplicity of the signals was established by the DEPT[9] pulse sequence. The characteristic chemical shift values of some of these signals[6-8,10] confirm the presence of two ester carbonyl groups, one trisubstituted carbon-carbon double bond and two carbon-carbon triple bonds, thus supporting the ideas inferred from IR, UV, ^1H NMR and MS data. Furthermore, four signals from oxygenated carbon atoms are visible in the range 60-80 ppm, and another signal appears at 103.49 ppm in the range of acetal carbons.



1 R = Me_2CHCH_2CO R' = H R" = OAc
2 R = Me_2CHCH_2CO R' = R" = H
3 R = R' = Ac R" = H

Scheme 1

Since only one olefinic signal is present, the signals at δ 5.99 (1H), 4.85 (1H) and 3.89 (3H) must arise from protons geminal to oxygen functions. Judging from the chemical shift, the three-proton multiplet centered at δ 3.9 has been assigned to hydrogens geminal to OH and ether oxygen atoms, whereas the two first signals correspond to protons geminal to acyloxy groups. One of them is an acetate group, which gives rise to the three-proton singlet at δ 2.07. The other, as deduced from the two methyl doublets in the vicinity of δ 1 and from the methylene signal (two double doublets) at δ 2.38 and 2.30, is an isovalerate (3-methylbutanoate) group. Both groups of signals are coupled to the methine complex signal at δ ca. 2.15 ppm, as established by spin decoupling.

The carbon-hydrogen connectivity was determined by a two-dimensional one-bond C-H shift correlation experiment.[9] While the ^1H signal at δ 5.99 correlates with the ^{13}C signal at 75.63 ppm, which in turn corresponds to an oxygenated carbon atom, the olefinic signal at δ 5.12 correlates with the carbon signal at 84.05 ppm. Such high field positions of olefinic carbon atoms have already been observed in the ^{13}C NMR spectra[6-8] of spiroacetal enol ethers. The other olefinic carbon appears, however, at a markedly low field (164.11 ppm), near the carbonyl range. The nonquaternary ^{13}C signals from the acetyl and isovaleroyl residues were also assigned by the C-H correlation.

The molecular formula indicates the existence of nine insaturations. The ester functions, the C=C and the two C≡C bonds account for seven these, the remaining two thus being assigned to rings. Furthermore, all seven oxygen atoms of the molecular formula are accounted for by the hydroxy group, the two ester functions and the acetal moiety. These facts indicate that the molecule of **1** contains a tetrahydrofurane/tetrahydropyrane spiroacetal unit bearing a hydroxy group, two ester residues and a C_6 diyne-ylidene chain (Scheme 1). Taking into account the chemical shifts of the signals and the results of the decoupling experiments, the hydroxy group has to be located at C-6, while the ester moieties are bound to C-2 and C-7.

The stereochemical features were deduced by NOE measurements. While most NOEs are straightforward and do not essentially contribute to the stereochemical elucidation, a significant one is observed between the signals of H-6 and the equatorial proton at C-4. In order to explain this NOE, it must be assumed that there is a syn relationship between H-6 and the methylene group (C-4) of the six-membered ring, taking the five-membered ring as reference (see Scheme 1). Furthermore, the absence of any measurable NOE between H-6 and H-7 suggests an *anti* relationship between both protons, thus corresponding with the probable biogenetic origin of **1** via *trans* opening of a spiroacetal epoxide.

The remaining structural aspect, the exact location of both ester residues, was determined with the aid of two-dimensional C-H correlations modulated by long-range couplings. Application of the COLOC[9] method permitted the observation of several correlations through two or three bonds, which are in agreement with the proposed structure. The most significant feature, however, is a weak, but detectable correlation between the acetyl carbonyl carbon and the signal from H-2 at δ 4.85. This allows the assignment of the acetoxyl group to C-2 and, consequently, the placement of the isovaleroyl residue at C-7. As represented in Scheme 1, however, structure **1** describes only the relative configuration of the molecule.

Table 1. ^{1}H-NMR data of compounds **1-3** *.

Compound	1	2	3	3#
Hydrogen				
H-1	3.88 br *d* (13) 3.96 *dd* (13; 1.5)	3.83 *m*+	3.82 *m*; 2H	3.56 *ddd* (13; 12; 2.5) 3.46 *ddd* (12; 5; 1.5)
H-2	4.85 br *s*	1.70-1.60 *m*; 2H	1.60-1.50 *m*; 2H	1.25 *m*; 1H 0.99 *m*; 1H
H-3	1.93 *m* 2.06 *m*	1.90-1.70 *m*; 4H	1.80-1.60 *m*; 4H	1.65-1.45 *m*; 4H
H-4	1.62 *ddd* (14; 4; 2) 2.15 *m*			
H-6	3.89 *d* (7)	3.83 *m*+	5.16 *d* (7.5)	5.42 *d*
H-7	5.99 *dd* (7; 2)	5.99 *dd* (7; 2)	6.25 *dd* (7.5; 2)	6.80 *dd*
H-9	5.12 *dq* (2; 1)	5.11 *dq* (2; 1)	5.14 *dq* (2; 1)	5.29 *dq*
H-14	1.93 *d* (1)	1.95 *d* (1)	1.96 *d* (1)	1.39 *d*
OAc	2.07 *s*; 3H	---	2.15 *s*; 6H	2.10 *s*; 3H 1.72 *s*; 3H
*i*Val	2.38 *dd*; 1H (15; 6.5) 2.30 *dd*; 1H (15; 6.5) 2.15 *m*; 1H 1.00 *d*; 3H (7) 0.98 *d*; 3H (7)	2.40 *dd*; 1H (15; 6.5) 2.31 *dd*; 1H (15; 6.5) 2.17 *m*; 1H 1.01 *d*; 3H (7) 0.99 *d*; 3H (7)	--- --- --- --- ---	--- --- --- --- ---

* At 400 MHz in $CDCl_3$ (25°C). Coupling constants (in Hz) are given in parentheses after the corresponding chemical shifts.
\# In C_6D_6. The coupling constants are the same as in $CDCl_3$; only those of H-1 are specifically shown.
\+ Overlapped.

The spectral data clearly suggest that the structure of compound **2** is closely related to that of **1**. Not only is the UV spectrum superimposable with that of **1**, but both IR spectra are also very similar. The exact mass of the molecular ion in the mass spectrum indicates the molecular formula $C_{19}H_{24}O_5$. In the ^{1}H NMR spectrum (Table 1), the signals of the terminal methyl group at δ 1.95 (d, J = 1 Hz), the olefinic signal at δ 5.11 (dq, J = 2 and 1 Hz) and the isovaleroyl residue are visible with practically the same position and shape as was observed in **1**. The most obvious difference is the absence of the acetyl singlet

in the vicinity of δ 2 ppm. Another significant feature is the absence of the acetoxy signals in the ^{13}C NMR spectrum, which is otherwise very similar to that of **1** (Table 2). These facts, together with the molecular formula, indicate that **2** has a structure analogous to **1**, in which a hydrogen atom replaces the acetate group at C-2.

Table 2. ^{13}C-NMR data of compounds **1-3** *

Compound	1	2	3
Carbon			
1	64.40	63.13	63.05
2	65.85	24.34	24.15
3	22.70	18.65	18.69
4	24.75	30.04	30.67
5	103.49	104.03	104.22
6	79.47	79.73	78.11
7	75.63	75.96	73.16
8	164.11	164.66	164.08
9	84.05	83.42	83.78
10	68.33	68.81	68.48
11	78.32	78.06	78.37
12	64.54	64.71	64.49
13	80.18	79.96	80.27
14	4.51	4.56	4.64
*i*Val	172.61	172.65	---
	42.56	42.67	---
	25.28	25.35	---
	22.48	22.54	---
	22.38	22.44	---
OAc	170.58	---	170.38; 169.87
	21.17	---	20.84; 20.41

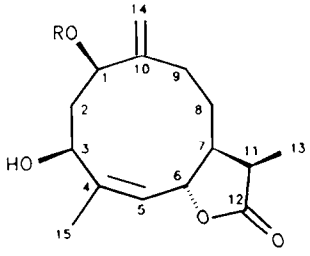
* At 67.89 MHz in $CDCl_3$ (25°C).

While compound **3** displays clear similarities to **1** and **2**, some differences may be noted. Despite the similarities in the UV spectra of the three products, and the fact that the molecular weight of **3**, deduced from MS data, is the same as that of **2**, it was found that the exact mass of the molecular ion corresponds to a different molecular formula, $C_{18}H_{20}O_6$. Another difference is the absence of hydroxyl bands in the IR spectrum, although bands from ester groups (1749 cm^{-1}) and acetylenic bonds (2230, 2139 cm^{-1}) are still visible. The ester carbonyl bands are due to two acetate groups, as evident in the NMR spectral data (Tables 1 and 2): a six-proton singlet at δ 2.15 ppm in the ^1H NMR spectrum and two sets of two peaks in the ^{13}C NMR spectrum in the ranges of about 21 and 170 ppm. In the mass spectrum, peaks due to the sequential loss of two acetic acid fragments are observed at m/z 272 and 212. Since all the other data are very close to those of the compounds discussed above, the suggested structure for compound **3** is one similar to that of **2**, but with two vicinal acetate residues at C-6 and C-7. NOE measurements not only confirmed this, but also clarified the structure's stereochemistry. In view of the overlapping of some key signals in $CDCl_3$, these measurements were performed in C_6D_6 (see Table 1). Saturation of the signal at δ 5.42 (H-6) gave rise to an NOE in part of the broad multiplet at 1.65-1.45, which can only correspond to H-3 and H-4. This reaffirms that H-6 is *syn* to the methylene chain of the tetrahydropyrane ring (see above). Further NOEs were observed between each of the acetate signals and *both* H-6 and H-7. This indicates a *trans* orientation of the acetate residues, since only thus can each acetate methyl group be spatially close to both hydrogen atoms. As in the former cases, structure **3** (Scheme 1) represents only the relative configuration of the molecule.

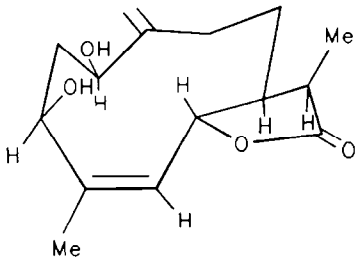
Compounds **4** and **5** are sesquiterpene lactones, as deduced from the IR carbonyl bands at about 1750 cm^{-1}, the molecular formulae, which contain fifteen carbon atoms, and the ^1H NMR data (Table 3). In addition to the lactone band, compound **4** displays a strong IR hydroxyl band. In the mass spectrum, a very weak molecular ion is visible at m/z 266, as well as two peaks corresponding to the loss of one and two water molecules, respectively. This suggests the presence of two hydroxyl groups. The exact mass measurement of the $[M^+-H_2O]$ ion (m/z 248.1412) yields a $C_{15}H_{20}O_3$ formula for this fragment, which renders the molecular formula $C_{15}H_{22}O_4$. This corresponds to the presence of five insaturations, two of them being assigned to the lactone part. Moreover, the lactone and the two hydroxyl groups account for the four oxygen atoms of the molecular formula.

The ^1H NMR spectrum indicates the presence of three olefinic hydrogens, two of them constituting a methylene unit (broad singlets at δ 5.35 and 5.15). This, plus the ^{13}C NMR data (Table 4), confirms the presence of a disubstituted and a trisubstituted double bond. The five insaturations only allow for the

existence of one ring. Indeed, the ^1H NMR spectrum of **4** resembles that of a germacranolide. More specifically, the presence of one methyl doublet at δ 1.17 (J = 7.5 Hz) is indicative of an 11,13-dihydrogermacranolide. Irradiation of this doublet (H-13, germacrane numbering, see Scheme 2) causes the collapse of a double quartet at δ 2.79 (J = 8.5 and 7.5 Hz) into a doublet with J = 8.5 Hz. This double quartet was thus assigned to H-11, which is coupled with H-13 and H-7. The size of the coupling constant $J_{7,11}$ suggests that both hydrogens are *cis* to each other, that is, that an 11αH configuration is present in this case.[11,12]



4 R = H
5 R = OH



4a

Scheme 2

Extensive decoupling experiments led to the establishment of the hydrogen connectivity and the assignment of the hydrogen signals. A three-proton doublet at δ 1.80 (J = 1.5 Hz) was assigned to an olefinic methyl group. Irradiation of this methyl signal transformed the signal at δ 5.36 (ddq, J = 8, 1.5 and 1.5) into a double doublet (J = 8 and 1.5 Hz), thus pointing to the olefinic nature of the latter. Three additional signals are visible in the low field range below 5 ppm. Two of them belong to the aforementioned methylene unit. The lowest field signal at δ 5.65 (J = 8 and 3 Hz) must therefore correspond to the proton geminal to the lactone oxygen (H-6). The observed coupling of this signal with those at 5.36 (olefinic, H-5) and 2.46 (H-7) confirmed this and the presence of a Δ^4 double bond. The sizes of the coupling constants of H-6 are similar to those commonly observed in 6,7-trans-heliangolides,[13-15] thus suggesting a trans lactone ring fusion and a cis stereochemistry for the double bond.

The broadened double doublets at δ 4.15 and 4.49 were assigned to hydrogens geminal to the two hydroxyl groups, the somewhat low-field δ values suggesting their allylic position. Irradiation of either signal caused alterations in the coupling pattern of two signals at δ 2.22 and 2.07. This clearly establishes the presence of the segment -CHOH-CH_2-CHOH-, which in turn allowed the assignment of both hydroxyl groups to C-1 and C-3 of the germacrane framework. Taking all these facts into account, an 11,13-dihydro-*trans*-germacran-12,6-olide structure with hydroxyl functions at C-1 and C-3, an exo-methylene unit at C-10 and a *cis* double bond at Δ^4 has been suggested for **4**.

The stereochemistry of compound **4** was determined by NOE measurements. Configurational and conformational assignments are always difficult in medium-sized rings, due to the high flexibility of such cyclic structures. In the case of germacrane derivatives, most studies[16-20] have focused on *trans,trans*-germacra-1(10),4-diene derivatives, the conformational preferences of *cis,trans*- or *cis,cis*-isomers being far less known. In the present case, an NOE was observed between the signal at 4.15 and one of the H-14 methylene singlets, indicating that this signal corresponds to H-1. Further NOEs are visible between H-1, H-3 and H-7, suggesting that all these hydrogens lie in the same (lower) face of the

medium ring surface. The hydroxyl groups at C-1 and C-3 are thus β-oriented. Of the further NOEs measured, those between the pairs H-6/H-13, H-7/H-11 and H-5/H-15 served to confirm the 11αH configuration at C-11 and the *cis* configuration of the double bond Δ^4.

Table 3. ^{1}H NMR data of compounds **4** and **5** *.

Compound	**4**	**5**
Hydrogen		
H-1	4.15 br *dd* (11, 3.5)	4.35 *dd* (11, 4.5)
H-2	2.22 *m* 2.07 *ddd* (15, 6.5, 3.5)	2.27 *m* (2H)
H-3	4.49 br *dd* (6.5, 2.5)	4.55 br *dd* (6.5, 2.5)
H-5	5.36 *ddq* (8, 1.5, 1.5)	5.39 *ddq* (8.5, 1.5, 1.5)
H-6	5.65 br *dd* (8, 3)	5.76 br *dd* (8.5, 3.5)
H-7	2.46 *dddd* (12, 8.5, 3, 3)	2.39 *dddd* (12, 9, 3.5, 3.5)
H-8	1.85 *m* 1.55 *m*	1.89 *m* 1.55 *m*
H-9	2.56 *ddd* (15, 9, 3) 2.22 *m*	2.58 *ddd* (15, 8, 3.5) 2.12 br *ddd* (15, 9, 4)
H-11	2.79 *dq* (8.5, 7.5)	2.81 *dq* (9, 7.5)
H-13	1.17 *d* (7.5)	1.18 *d* (7.5)
H-14	5.35 br *s* 5.15 br *s*	5.46 br *s* 5.37 br *s*
H-15	1.80 *d* (1.5)	1.80 *d* (1.5)
Other signals		7.80 br *s* (OOH)

* At 400 MHz in $CDCl_3$ (25°C). Coupling constants (in Hz) are given in parentheses after the corresponding chemical shifts.

Table 4. ^{13}C-NMR data of compounds **4** and **5** *.

Compound	**4**	**5**
Carbon		
1	73.27	86.07
2	39.26	34.52
3	70.95	71.36
4	142.32	142.20
5	124.80	125.41
6	78.46	78.45
7	42.06	43.22
8	27.29	27.67
9	29.18	31.29
10	149.66	144.75
11	37.49	37.62
12	179.41	179.46
13	10.32	10.59
14	115.25	118.80
15	22.66	22.84

* At 67.89 MHz in $CDCl_3$ (25°C).

Compound **4** is one of the two epimeric 11,13-dihydroderivatives of pulverolide, a sesquiterpene lactone isolated from *Leucanthemopsis pulverulenta*.[14] The other epimer at C-11 has recently been reported in *Pyrethrum santolinoides*.[21] The NOEs between H-5/H-7, H-1/H-15, H-3/H-15 suggest a predominant conformation with H-5 and H-15 pointing downwards, similar to **4a** (Scheme 2). The low-field position of H-6 is probably due to its spatial proximity with the 3β-OH group.

The spectral features of compound **5** are very similar to those of **4**. The ^{1}H NMR spectra, for instance, clearly resemble each other (Table 3). The most significant difference is that one signal at δ 4.35, which appears with practically the same shape as the signal of H-1 in **4**, is shifted 0.2 ppm to lower field. The broad singlets of the exocyclic methylene protons have also undergone similar low-field shifts of about 0.2 ppm (Table 3). A very broad singlet in the ^{1}H NMR spectrum at δ 7.80 suggests the presence of an hydroperoxide. Since the other signals appear more or less in the same position in the spectra of both compounds, the most reasonable proposal for the structure of **5** is one identical to that of **4** with a hydroperoxy group replacing the allylic OH at C-1. This proposal was strengthened by the fact that reduction with triphenylphosphine transformed **5** into **4**. A compound closely related to **5** has recently been isolated from *Anthemis nobilis*.[22] Its ^{1}H and ^{13}C NMR spectral data are in part very similar to those of **5**, giving further evidence in favour of the proposed structure.

Aside from the new compounds described above, some other known products were also found in *C. lavandulifolium*. They were identified by their spectral properties with the guaianolides kauniolide,[23]

dehydroleucodin,[24] cumambrin A,[25] tanaparthin-α-peroxide,[26,27] canin,[26,27] the 1,10-secoguaianolide secotanapartholide A,[26,27] and the dimeric guaianolide handelin.[28,29]

EXPERIMENTAL

NMR spectra were measured on Bruker NMR spectrometers WM-400 and WM-270 at the frequencies indicated in the Tables. Two-dimensional correlation spectra were measured with standard Bruker software. Mass spectra were run on a Varian MAT 711 spectrometer. IR spectra (oily films) were recorded on a Perkin Elmer IR spectrophotometer mod. 281. HPLC was performed in the reverse phase mode (column 1: LiChrosorb RP-8, 250 x 25 mm, flow = 12 mL/min; column 2: LiChrosorb RP-8, 250 x 8 mm, flow = 3 mL/min; elution with MeOH-H_2O mixtures). Medium pressure column chromatography (MPCC) was made on silica gel Woelm (40-63 μ).

Extraction and chromatography: *Chrysanthemum lavandulifolium* was collected in October 1986 in Myohyangsan, People's Republic of Korea, and authenticated by Prof. Kim Hjon-Sam. Aerial parts of the plant (300 g) were air-dried at room temperature, ground and extracted at room temp. with hexane-Et_2O-MeOH 1:1:1.[30] The extract (14.6 g) was defatted by precipitation from MeOH (150 mL) at -15° and the material obtained (9 g) was prefractionated by column chromatography on silica gel: A, hexane-Et_2O 3:1; B, hexane-Et_2O 1:1; C, hexane-Et_2O 1:3, D, Et_2O and E, Et_2O-MeOH 9:1 (length, 70 cm, i.d. 5 cm, 4 L of each solvent mixture). These five fractions were subjected to diverse chromatographic separations as described below. The fractions obtained in the successive separation phases were further processed or rejected according to their weight and complexity, as judged by TLC and ^1H NMR.

Fraction A consisted mainly of waxes, volatile terpenes and sterols (^1H NMR). MPCC of this fraction (elution with hexane-Et_2O 9:1 to 1:1) gave kauniolide (12 mg). Fraction B was submitted to HPLC (column 1, elution with MeOH-H_2O 75:25). This yielded two crude fractions, which were further purified by repeated preparative TLC (elution with hexane-Et_2O 1:1), affording **2** (28 mg) and **3** (11 mg). HPLC of fraction C (column 1, elution with MeOH-H_2O 75:25) gave fractions C-1 and C-2. Fraction C-1 yielded tanaparthin-α-peroxide (15 mg) after preparative TLC (elution with hexane-Et_2O 1:2). Fraction C-2 contained only **1** (108 mg). MPCC of fraction D (elution with $CHCl_3$-MeOH 100:1 to 25:1) gave fractions D-1 to D-3. Fraction D-1 was submitted to preparative TLC (elution with hexane-Et_2O 1:3). This gave dehydroleucodin (10 mg) and cumambrin A (20 mg). Fraction D-2 contained only cumambrin A (100 mg). Fraction D-3 was further purified by preparative TLC (elution with Et_2O) and HPLC (column 2, elution with MeOH-H_2O 55:45). This gave lactone **5** (5 mg).

Fraction E was first fractionated by MPCC (elution with $CHCl_3$-MeOH 25:1) to three fractions, E-1 to E-3. Fraction E-1 deposited crystals of handelin (40 mg) by standing. The mother liquors were submitted to HPLC (column 2, elution with MeOH-H_2O 1:1) and then preparative TLC (elution with $CHCl_3$-MeOH 25:1). This gave secotanapartholide A (10 mg). Fraction E-2 was purified by HPLC (column 2, elution with MeOH-H_2O 55:45), affording canin (8 mg). Finally, fraction E-3 was further purified by preparative TLC (elution with $CHCl_3$-MeOH 25:1) and then HPLC (column 2, elution with MeOH-H_2O 55:45). This gave lactone **4** (12 mg).

(+)-(3S*,4S*,5R*,8S*)-(E)-8-*Acetoxy*-4-*hydroxy*-3-*isovaleroyloxy*-2-*(hexa*-2,4-*diynyliden)*-1,6-*dioxaspiro*[4,5]*decane* (**1**). Yellowish oil, $[\alpha]_D^{24}$ +62° (c, 5.66; $CHCl_3$). IR ν max (film): 3450 (br, OH), 2229, 2139 (C≡C), 1733 (ester C=O), 1650 (C=C), 1370, 1232, 1175, 1012, 958, 885 cm^{-1}. UV λ max (MeOH): 216, 224, 250sh, 263, 276, 291 nm. EIMS m/z (% rel. int.): 390 (M^+, 45), 372 (M^+-H_2O, 5), 332 (4), 306 (M^+-C_5H_8O, 2), 288 (M^+-$C_5H_{10}O_2$, 3), 246 (8), 199 (30), 136 (96), 124 (77), 121 (58), 111 (100), 104 (88). Exact mass measurement for the molecular ion: found, M = 390.1686; calcd. for $C_{21}H_{26}O_7$: M = 390.1679. For NMR data, see Tables 1 and 2.

(+)-(3S*,4S*,5R*)-(E)-4-*hydroxy*-3-*isovaleroyloxy*-2-*(hexa*-2,4-*diynyliden)*-1,6-*dioxaspiro*[4,5]*decane* (**2**). Yellowish oil, $[\alpha]_D^{24}$ +68° (c, 2.28; $CHCl_3$). IR ν max (film): 3460 (br, OH), 2231, 2139 (C≡C), 1743 (ester C=O), 1645 (C=C), 1370, 965, 935, 880 cm^{-1}. UV λ max (MeOH): 216, 224, 250sh, 263, 277, 292 nm. EIMS m/z (% rel. int.): 332 (M^+, 8), 314 (M^+-H_2O, 1), 272 (2), 230 (M^+-H_2O-C_5H_8O, 18), 288 (M^+-$C_5H_{10}O_2$, 3), 246 (8), 199 (30), 136 (96), 220 (16), 136 (31), 126 (40), 124 (85), 85 (71), 57 (100). Exact mass measurement for the molecular ion: found, M = 332.1623; calcd. for $C_{19}H_{24}O_5$: M = 332.1624. For NMR data, see Tables 1 and 2.

(−)-(3S*,4S*,5R*)-(E)-3,4-*Diacetoxy*-2-*(hexa*-2,4-*diynyliden)*-1,6-*dioxaspiro*[4,5]*decane* (**3**). Yellowish oil, $[\alpha]_D^{24}$ -1° (c, 1.16; $CHCl_3$). IR ν max (film): 2230, 2139 (C≡C), 1749 (ester C=O), 1651 (C=C), 1215, 875, 810 cm^{-1}. UV λ max (MeOH): 216, 223, 250sh, 262, 276, 291 nm. EIMS m/z (% rel. int.): 332 (M^+, 6), 272 (M^+-HOAc, 8), 230 (M^+-HOAc-C_2H_2O, 41), 212 (10), 202 (10), 168 (25), 126 (100), 71 (40). Exact mass measurement for the molecular ion: found, M = 332.1267; calcd. for $C_{18}H_2O_6$: M = 332.1260. For NMR data, see Tables 1 and 2.

1β,3β-*Dihydroxygermacra*-4Z,10(14)-*dien*-6β,7α,11αH-12,6-*olide* (**4**). Colourless gum, $[\alpha]_D^{24}$ +26° (c, 0.9; $CHCl_3$). IR ν max (film): 3400 (br, OH), 1748 (lactone C=O), 1640, 1435, 1375, 1195, 1175, 1025 cm^{-1}. EIMS m/z (% rel. int.): 266 (M^+, 1), 248 (M^+-H_2O, 2), 233 (M^+-H_2O-Me, 1), 230 (M^+-$2H_2O$, 1), 205 (3), 193 (7), 181 (7), 175 (8), 120 (58), 105 (100), 95 (39), 91 (60), 84 (98). Exact mass measurement for [M^+-H_2O]: M = 248.1412. Calc. for $C_{15}H_{20}O_3$, M = 248.1412. For NMR data, see Tables 3 and 4.

1β-*Hydroperoxy*-3β-*hydroxygermacra*-4Z,10(14)-*dien*-6β,7α,11αH-12,6-*olide* (**5**). Colourless gum. IR ν max (film): 3400 (br, OH), 1750 (lactone C=O), 1659, 1440, 1370, 1190, 950, 730 cm^{-1}. EIMS m/z (% rel. int.): 264 (M^+-H_2O, 3), 248 (M^+-H_2O_2, 3), 233 (M^+-H_2O_2-Me, 6), 191 (20), 175 (21), 135 (44), 121 (66), 107 (81), 95 (100), 69 (59). For NMR data, see Tables 3 and 4. Reaction of **5** with triphenylphosphine in CH_2Cl_2 at room temperature (1 hr) yields **4**.

All known compounds were identified by comparison with authentic samples from either our or Prof. Bohlmann's collection.

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