# Structure of Conocephalenol, a Sesquiterpenoid Alcohol from the European Liverwort *Conocephalum conicum*: Determination of the Absolute Configuration by Total Synthesis

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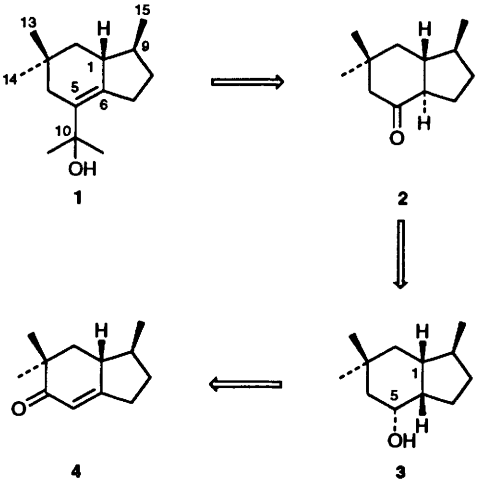
The structure of conocephalenol, a brasilane sesquiterpenoid alcohol isolated from the European liverwort *Conocephalum conicum*, has been determined by extensive NMR studies including a 2D INADEQUATE experiment and by total synthesis, which established the absolute stereochemistry as 1*R*,9*S*.

Liverworts are rich sources of terpenoids with unusual frameworks.[1] Such metabolites often exhibit interesting biological properties.[1] *Conocephalum conicum* is a common thalloid liverwort which occurs abundantly. A previous investigation of *C. conicum* resulted in the identification of methyl cinnamate, oct-1-en-3-ol and its acetate, which are responsible for the mushroom odour of this liverwort.[1] The diethyl ether or hexane extracts of the species collected in Scotland or Germany showed a red spot on TLC after spraying with sulfuric acid and heating. Extracts of species collected in Japan did not show such a spot. The compound which gives rise to this spot is the sesquiterpenoid alcohol conocephalenol **1**. We now report details of the structural elucidation of conocephalenol and of its total chiral synthesis which establishes its absolute stereochemistry as 1*R*,9*S*.

## Results and Discussion

*C. conicum* was collected in Scotland[2] or in Germany and was extracted with diethyl ether or hexane. Repeated chromatographic separation afforded a sesquiterpenoid alcohol which we named conocephalenol **1**, $[\alpha]_D^{22}$ −4.77[†] (*c* 1.30, EtOH). The molecular formula $C_{15}H_{26}O$ was determined by high-resolution mass spectrometry (HRMS). The IR spectrum showed the presence of a hydroxy group, which the $^{13}C$ NMR spectrum [$\delta_C$ 74.0 (s)] showed to be tertiary. The $^1H$ and $^{13}C$ spectra revealed two sets of *gem*-dimethyl groups [$\delta_H$ 1.25 (6 H), 1.00 (3 H) and 0.90 (3 H)], a secondary methyl group [$\delta_H$ 1.01 (d, *J* 7.3 Hz)] and a tetrasubstituted double bond ($\delta_C$ 135.9 and 132.7). The molecule is therefore bicarbocyclic. It was not possible to obtain any connectivity data from the $^1H$ NMR spectrum of conocephalenol because of its congested nature. Fortunately, we isolated a sufficient amount of conocephalenol **1** to allow us to perform a 2D INADEQUATE experiment, which clearly revealed the brasilane skeleton of conocephalenol as in structure **1**. This unusual carbon skeleton has previously been reported for brasilenol,[3] a marine natural product. Conocephalenol represents the first example of the isolation of a compound of this class from a liverwort. This finding may indicate a common ancestry for liverworts and algae.[1] Unfortunately, it was not possible to derive the relative stereochemistry of the molecule because of severe signal overlaps in the $^1H$ NMR spectrum. The relative stereochemistry was eventually assigned as 1*R**,9*S** as in structure **1** by independent syntheses.[2,4] The limited availability of conocephalenol and its relative instability under acidic conditions precluded a chemical approach to the determination of the absolute configuration and so we turned our attention to a chiral synthesis.

The retro-synthetic plan is shown in Scheme 1. We selected as starting material the trimethylhydrindenone **4**, previously used in the Tokushima racemic synthesis.[4] Reduction and 1,2-carbonyl transposition, followed by resolution of the camphanoyl ester of the intermediate alcohol **3** and subsequent alkylation should lead to the natural product (or its antipode).



**Scheme 1** Synthetic plan for conocephalenol **1**

The enone **4**[5] was hydrogenated (Scheme 2) in the presence of 10% Pd–C to afford the *cis*-hydrindanone **5**,[4] which was reduced by $NaBH_4$, followed by dehydration with $POCl_3$, to give the olefin **7**. Hydroboration–oxidation afforded a mixture of the 5α-ol **3** [$\delta_H$ 4.11 (dt, *J* 11.7 and 5.7, 5-H)] and the 4-ols **6**.[‡] The latter were recycled to yield more 5α-ol **3**. The coupling constants of 5-H reveal the equatorial (α) nature of the hydroxy group. The alcohol **3** was treated with (1*S*)-(−)-camphanoyl chloride and 4-(dimethylamino)pyridine (DMAP) in pyridine–dichloromethane. The diastereoisomeric products were separated by HPLC (Develosil 60-10) to afford compounds (+)-**8** and (−)-**9**. The absolute configuration was deduced from the CD spectrum of the ketone derived from ester **9**. Compound **9** was treated with 5% KOH–MeOH to give the corresponding alcohol (−)-**3** (Scheme 4), which was subjected to Jones

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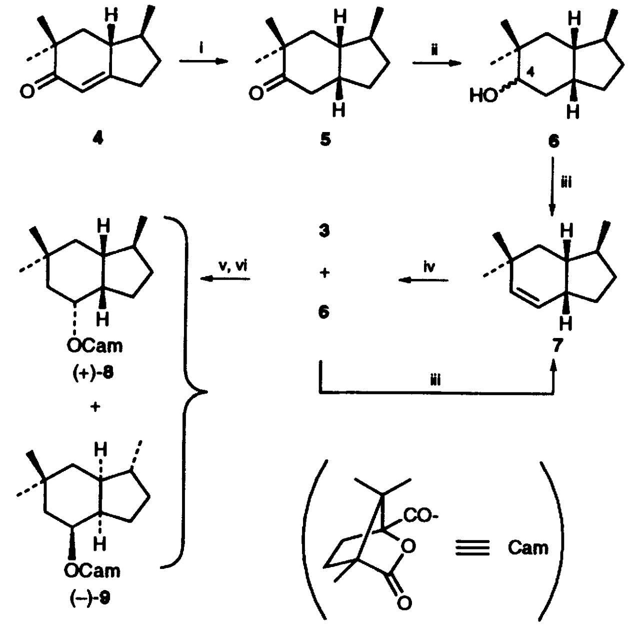
[†] Throughout, specific optical rotations $[\alpha]_D$ are given in units of $10^{-1}$ deg cm$^2$ g$^{-1}$.

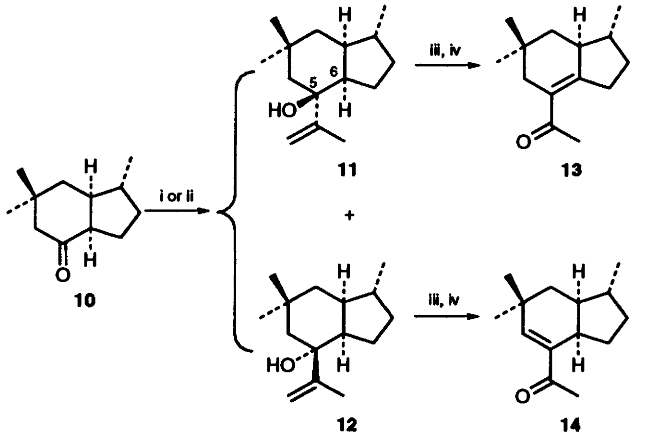
[‡] The numbering system for brasilenol[3] is used.



Scheme 2 *Reagents:* i, $H_2$/Pd–C, MeOH; ii, $NaBH_4$, MeOH; iii, $POCl_3$, Py; iv, $BH_3$; then $H_2O_2$; v, (1*S*)-(−)-camphanoyl chloride, DMAP, Py–$CH_2Cl_2$; vi, HPLC, Develosil 60-10, hexane–EtOAc

oxidation to give ketone (−)-**10**. The CD spectrum of this ketone showed a negative Cotton effect, which leads to the absolute configuration shown, on the basis of the octant rule (Fig. 1).

Conditions for the alkylation step (Scheme 3) were



Scheme 3 *Reagents and conditions:* i, Mg, 2-bromopropene, THF (45% yield; **11**:**12** 1:3); ii, Bu$^t$Li, 2-bromopropene, THF, −78 °C (20% yield; **11** only); iii, $O_3$; $Me_2S$; iv, $SOCl_2$

established by using the racemic ketone **10**. Alkylation with isopropenylmagnesium bromide in tetrahydrofuran (THF) afforded alcohols **11** and **12** in the ratio 1:3 (45% yield). The minor alcohol **11** gave the tetrasubstituted enone **13** in three steps. The observation of a nuclear Overhauser enhancement (NOE) between a methyl group at C-3 and the isopropenyl group in structure **11** indicated that these groups are axial and thus the hydroxy group at C-5 is *trans* to 6-H. Meanwhile the major isomer **12** was converted into the trisubstituted enone **14**. The yield in the alkylation step was very poor. Reaction of ketone **10** with isopropenyllithium at −78 °C produced the desired alcohol **11** as the sole product, but in very low yield (~20%). This approach was therefore abandoned in favour of alkylation of the *trans*-ketone **2** (Scheme 4).

The *cis*-ketone (−)-**10** was exposed to isomerization conditions ($K_2CO_3$, MeOH, reflux) to yield the *trans*-ketone (−)-**2**. Unchanged *cis*-ketone (−)-**10** was recovered by HPLC and recycled to produce more *trans*-ketone (−)-**2**. Alkylation of ketone (−)-**2** should occur from the β face of the molecule because of the steric effect of the axial methyl group at C-3. The product should be the axial alcohol **15**. In the event, alkylation of *trans*-ketone (−)-**2** with isopropenyllithium in THF at −78 °C afforded the axial alcohol (−)-**15** as the sole product and in excellent yield (91%). This was converted as above, *via* the acyloin (−)-**16** (Scheme 4), into the desired tetrasubstituted enone (−)-**13**, which was methylated (MeLi) to give (+)-conocephalenol (+)-**1**, $[\alpha]_D^{22}$ +5.85 (*c* 0.94), identical in all respects with natural conocephalenol apart from the sign of rotation. Thus the absolute configuration of natural conocephalenol (−)-**1** is 1*R*,9*S*.

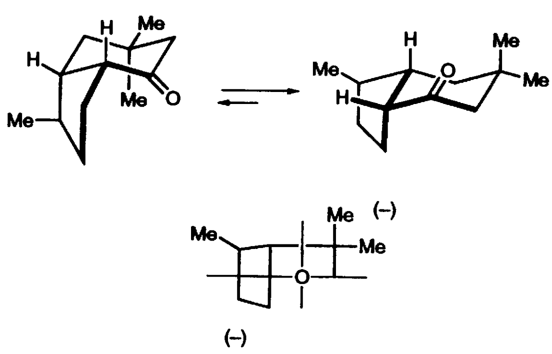
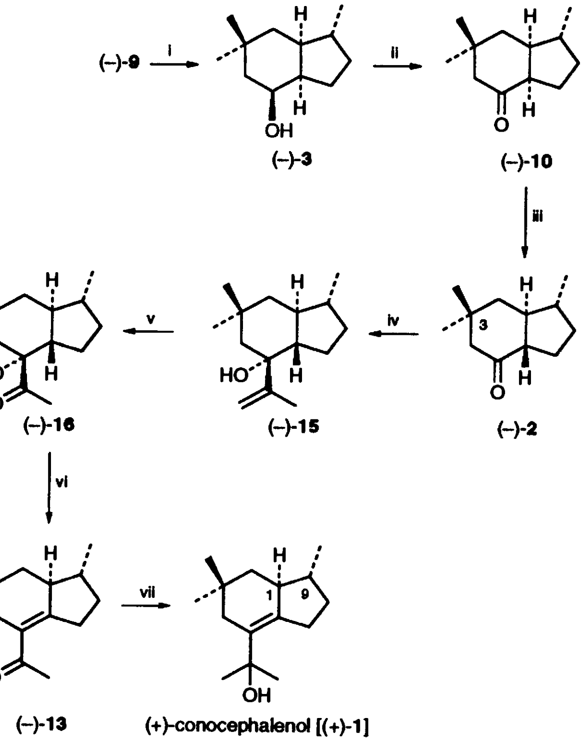


**Fig. 1** Conformations of the ketone (−)-**10** and back octant



Scheme 4 *Reagents:* i, KOH, MeOH; ii, Jones oxidation; iii, $K_2CO_3$, MeOH; iv, Bu$^t$Li, 2-bromopropene, THF; v, $O_3$; then $Me_2S$; vi, $SOCl_2$, Py; vii, MeLi

## Experimental

*General.*—IR spectra were measured on a Shimadzu IR-408 spectrophotometer. $^1H$ and $^{13}C$ NMR spectra were recorded on a JEOL JNM GX-400, Bruker WP200SY, Varian Unity 200 or FX-90Q spectrometer. The solvent used for NMR spectra was

$CDCl_3$ unless otherwise stated. *J*-Values are given in Hz. Mass spectra were measured on a JEOL JMS HX-100 or AX-500 spectrometer. The specific rotations and the CD spectra were taken on a JASCO DIP-140 polarimeter and a JASCO J-500 spectrometer, respectively. Chemcopak Nucleosil 50-5 (10 × 250 mm), Develosil 60-3 (4.6 × 150 mm), Develosil 60-10 (20 × 250 mm) or Cosmosil 5C18 (10 × 250 mm) columns were used for HPLC (JASCO pump system). Silica gel 60 for column chromatography was purchased from Merck.

*Isolation.*—The liverwort *C. conicum* was collected in Scotland in 1986 and in Germany in July 1991. The dried German material (237 g) was pulverized and extracted with hexane to afford a residue (4.5 g). The extract (2.2 g) was subjected to Sephadex LH-20 column chromatography (elution with MeOH) twice to afford a conocephalenol-containing faction (786 mg). This fraction was further separated by reversed-phase HPLC [Cosmosil 5C18; elution with MeOH–water (9:1)] to give a conocephalenol-containing fraction (145 mg). Then finally the mixture was purified by normal-phase HPLC (Develosil 60-3; hexane–EtOAc (9:1)] to afford pure *conocephalenol* **1** (13 mg) as an oil; $[\alpha]_D^{22}$ −4.77 (*c* 1.3, EtOH); $\nu_{max}$(film)/cm$^{-1}$ 3450; $\delta_H$(400 MHz; $C_6D_6$)§ 0.90 (3 H, s), 1.00 (3 H, s), 1.01 (3 H, d, *J* 6.4) and 1.25 (6 H, s); $\delta_C$(100 MHz; $C_6D_6$)§ 18.1, 26.6, 29.3, 29.5, 29.99, 30.04, 32.5, 34.1, 40.5, 40.8, 41.5, 48.3, 74.0, 132.7 and 135.9; *m/z* 222 ($M^+$), 204, 189, 175, 161, 149, 133, 127, 119, 105, 91, 84, 69, 59 (base) and 55 (Found: $M^+$, 222.2009. $C_{15}H_{26}O$ requires M, 222.1984). The yields of conocephalenol from Scottish *C. conicum* were much higher but very dependent on the time between collection and extraction.

*Synthesis.*—*Hydrogenation of the trimethylhydrindenone* **4**. The enone **4** (2.4 g) was hydrogenated in the presence of 10% Pd–C (200 mg) in MeOH (50 cm$^3$). The solvent was evaporated off after filtration of the catalyst to afford the ketone **5** (2.3 g, 95%),[4] oil; $\nu_{max}$(film)/cm$^{-1}$ 1705; $\delta_H$(200 MHz) 0.94 (3 H, d, *J* 6.5), 1.05 (3 H, s) and 1.09 (3 H, s); $\delta_C$(50 MHz) 18.8 (Me), 23.7 (Me), 25.9 (Me), 32.8 ($CH_2$), 34.1 ($CH_2$), 36.0 (CH), 40.8 ($CH_2$), 41.4 (CH), 41.6 (CH), 42.3 (C), 42.5 ($CH_2$), and 216.7 (CO); *m/z* 180 ($M^+$), 162, 151, 147, 136 (base), 121, 109, 95, 86, 81, 70 and 55.

*Preparation of the olefin* **7**. A solution of ketone **5** (4.1 g) in MeOH (50 cm$^3$) was treated with $NaBH_4$ (1 g) at room temperature for 2.5 h. Hydrochloric acid (1 mol dm$^{-3}$) was added and MeOH was evaporated off under reduced pressure. The residue was extracted with diethyl ether and the ethereal solution was washed with brine. Evaporation of the solvent, after drying over $MgSO_4$, afforded the alcohol **6** (3.97 g, 97%).

To a solution of the alcohol **6** (4.86 g) in pyridine (30 cm$^3$) was added $POCl_3$ (6 cm$^3$) and the mixture was stirred at room temperature overnight. Water was added and the mixture was extracted with diethyl ether. The ethereal solution was washed successively with 1 mol dm$^{-3}$ HCl, 5% aq. $NaHCO_3$ and brine, dried over $MgSO_4$, and concentrated to give the *olefin* **7** (3.97 g, 91%) as an oil; $\nu_{max}$(film)/cm$^{-1}$ 1635, 1450, 1370 and 1360; $\delta_H$(200 MHz) 0.95 (3 H, s), 0.97 (3 H, s), 1.02 (3 H, d, *J* 6.8), 5.43 (1 H, dd, *J* 9.7 and 1.7) and 5.59 (1 H, dd, *J* 9.9 and 3.9); $\delta_C$(50 MHz) 21.3 (Me), 27.7 (Me), 31.1 (Me), 32.3 (C), 32.7 ($CH_2$), 34.4 ($CH_2$), 38.7 (CH), 41.3 (CH), 41.9 (CH), 42.0 ($CH_2$), 127.8 (CH) and 136.8 (CH); *m/z* 164 ($M^+$), 149 (base), 135, 121, 108, 107, 93, 79, 67, 53 and 41 (Found: $M^+$, 164.1568. $C_{12}H_{20}$ requires M, 164.1566).

*Hydroboration–oxidation of the olefin* **7**. To a solution of olefin **7** (5.56 g) in dry THF (90 cm$^3$) was added 1 mol dm$^{-3}$ $BH_3$·THF (79 cm$^3$) and the mixture was stirred at room temperature for 2 h. Water (14.5 cm$^3$), 3 mol dm$^{-3}$ NaOH (30 cm$^3$) and 30% $H_2O_2$ (30 cm$^3$) were added and the mixture was stirred for 1 h. The mixture was treated with 10% aq. $Na_2S_2O_3$ for 0.5 h, and THF was evaporated off. The residue was extracted with diethyl ether and the organic phase was washed with brine, dried over $MgSO_4$, and evaporated to afford a residue (6.06 g), which was subjected to silica gel column chromatography (400 g) and elution with hexane–EtOAc gradient to afford the alcohol **3** (1.10 g, 18%) and its regioisomer **6** (1.86 g, 30%). *Alcohol* **3**; gum; $\nu_{max}$(film)/cm$^{-1}$ 3600 and 3425; $\delta_H$(200 MHz) 0.88 (3 H, s), 0.91 (3 H, s), 0.95 (3 H, d, *J* 7.0) and 4.11 (1 H, dt, *J* 11.7 and 5.7); $\delta_C$(50 MHz) 21.9 ($CH_2$), 22.7 (Me), 24.6 (Me), 30.7 ($CH_2$), 32.4 (C), 33.1 (Me), 39.3 (CH), 41.1 ($CH_2$), 42.2 ($CH_2$), 42.9 (CH), 44.3 (CH) and 68.5 (CH); *m/z* 182 ($M^+$), 164, 149, 135, 125, 121, 108, 95, 93 (base), 85, 81, 79, 69, 67, 55 and 41 (Found: $M^+$, 182.1682. $C_{12}H_{22}O$ requires M, 182.1671).

*Reaction of the alcohol* **3** *with* (1S)-(−)-*camphanoyl chloride*. A solution of the alcohol **3** (1.51 g) in dry dichloromethane (50 cm$^3$) was treated with (1*S*)-(−)-camphanoyl chloride (3.5 g), pyridine (3 cm$^3$) and DMAP (600 mg) at room temperature overnight. Water was added and the mixture was extracted with diethyl ether. The ethereal solution was washed successively with 1 mol dm$^{-3}$ HCl, 5% aq. $NaHCO_3$ and brine, dried over $MgSO_4$, and evaporated to afford a residue (3.02 g). The residue was separated by HPLC [Develosil 60-10; elution with hexane–benzene–EtOAc (4.5:4.5:1), flow rate 7.5 cm$^3$ min$^{-1}$] to give the esters (+)-**8** (1.15 g, 38%) and (−)-**9** (1.09 g, 36%).

*Ester* (+)-**8**; mp 145–148 °C (from hexane); $[\alpha]_D^{23}$ +13.6 (*c* 1.03, $CHCl_3$); $\nu_{max}$($CHCl_3$)/cm$^{-1}$ 1780, 1730, 1710, 1270 and 1105; $\delta_H$(400 MHz) 0.947 (3 H, s), 0.95 (3 H, d, *J* 7), 0.97 (3 H, s), 1.05 (3 H, s), 1.11 (6 H, s) and 5.39 (1 H, ddd, *J* 17, 11 and 6.4); $\delta_C$(22.5 MHz) 9.5 (Me), 16.6 (Me), 16.7 (Me), 22.4 (Me), 23.1 ($CH_2$), 24.3 (Me), 28.9 ($CH_2$), 30.5 ($CH_2$ × 2), 32.3 (C), 32.7 (Me), 38.4 ($CH_2$), 39.3 (CH), 39.5 (CH), 40.8 ($CH_2$), 44.1 (CH), 53.7 (C), 54.6 (C), 73.8 (CH), 91.0 (C), 166.6 (C) and 177.9 (C); *m/z* (CI, isobutane) *m/z* 363 (M + H)$^+$, 279, 239, 199, 181, 165 (base), 109, 95 and 69 [Found (CI, isobutane): $M^+$, 363.2538 (M + H)$^+$. $C_{22}H_{35}O_4$ requires *m/z*, 363.2535].

*Ester* (−)-**9**; mp 121–122 °C (from hexane); $[\alpha]_D^{23}$ −30.1 (*c* 1.08, $CHCl_3$); $\nu_{max}$($CHCl_3$)/cm$^{-1}$ 1780, 1730 and 1715; $\delta_H$(400 MHz) 0.97–0.93 (12 H), 1.05 (3 H, s), 1.11 (3 H, s) and 5.36 (1 H, dt, *J* 11.0 and 5.8); $\delta_C$(50 MHz) 9.60 (Me), 16.7 (Me × 2), 22.5 (Me), 23.2 ($CH_2$), 24.3 (Me), 28.8 ($CH_2$), 30.6 ($CH_2$ × 2), 32.3 (C), 32.8 (Me), 38.2 ($CH_2$), 39.3 (CH), 39.4 (CH), 40.8 ($CH_2$), 44.1 (CH), 53.6 (C), 54.6 (C), 74.0 (CH), 91.0 (C), 166.7 (C=O) and 178.0 (C=O); *m/z* (CI, isobutane) 363 (M + H)$^+$, 199, 181, 164 (base), 149, 109, 95, 83, 69 and 55 [Found (CI, isobutane): 363.2534 (M + H)$^+$. $C_{22}H_{35}O_4$ requires *m/z*, 363.2535].

*Hydrolysis of the camphanoyl ester* (−)-**9**. The ester (−)-**9** (1.09 g) was treated with 5% KOH–MeOH (25 cm$^3$) at room temperature overnight. Hydrochloric acid (1 mol dm$^{-3}$) was added, methanol was evaporated off, and the residue was extracted with diethyl ether. The ethereal solution was washed successively with 5% aq. $NaHCO_3$ (3 ×) and brine, and dried over $MgSO_4$. Evaporation of the solvent afforded an alcohol (−)-**3** (471 mg, 86%), mp 65–70 °C (from $Et_2O$); $[\alpha]_D^{23}$ −33.3 (*c* 1.16, $CHCl_3$).

*Jones oxidation of the alcohol* (−)-**3**. A solution of the alcohol (−)-**3** (471 mg) in acetone (20 cm$^3$) was treated with Jones' reagent (8 mol dm$^{-3}$; 1.5 cm$^3$) at 0 °C and the mixture was stirred for 15 min. Isopropyl alcohol was added, acetone was removed, and the residue was extracted with diethyl ether. The ethereal solution was washed successively with 5% aq. $NaHCO_3$ and brine, dried over $MgSO_4$, and evaporated to afford the *ketone* (−)-**10** (453 mg, 97%) as an oil; $[\alpha]_D^{24}$ −101.7 (*c* 1.50, EtOH); $\nu_{max}$(film)/cm$^{-1}$ 1700; $\delta_H$(200 MHz; $C_6D_6$) 0.74 (3 H, s),

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§ Since conocephalenol **1** rapidly decomposes in $CDCl_3$ solution, the NMR spectra were taken in $C_6D_6$.

($M^+$), 207, 204, 189, 175, 164, 149, 133, 119, 107, 93, 79, 59 (base) and 55.

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