between the molecule halves. Solvent molecules were not included in the calculations, but the effect was taken into account by employing a dielectric constant of 5.0 for $CHCl_3$ and 78.0 for $(CH_3)_2SO$;[22] the value for $(CH_3)_2SO$ corresponds to that for H_2O.

The starting conformations for energy minimization calculations were built up by using the possible torsion angles estimated by NMR experiments. Any structural constraints imposed while building plausible starting conformations were relaxed during the calculation, and the energy minimization was allowed to proceed freely without any conformational constraints. The refined conformers that were not consistent with the NMR results were rejected from the consideration.

Registry No. 1, 74847-09-9.

(22) Weast, R. C.; Astle, M. J.; Beyer, W. H. *CRC Handbook of Chemistry and Physics*, 69th ed.; CRC Press: Boca Raton, FL, 1988.

Structure and Absolute Configuration of Spathulasin, a Metabolite of *Aeonium spathulatum*

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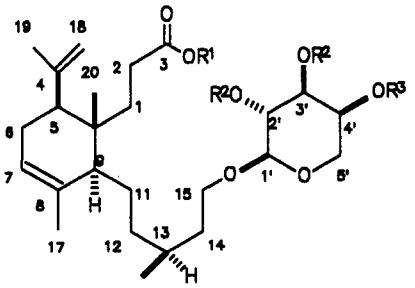
A new metabolite, spathulasin (1), is described as a natural product from *Aeonium spathulatum*. The structure of the 14-membered lactone derivative **10** was determined by X-ray crystallographic analysis. Hydrolysis of spathulasin and identification of the pentose portion as L-arabinose allowed the assignation of the absolute configuration of all centers in **1**.

Among the genera of Crassulaceae, *Aeonium*, which is developed to a striking degree in the Canary Islands,[1] has received scarce attention from the chemical point of view.[2] We have investigated the constituents of *Aeonium spathulatum* (Hornem.) Praeger and herein describe the isolation and structure determination of a novel diterpene glycoside.

Spathulasin (1) was isolated from the alcoholic extract of *A. spathulatum* as an amorphous solid. Data from



1 $R^1 = R^2 = H$, $R^3 = (CH_3)_2C{=}CHCO$

2 $R^1 = Me$, $R^2 = H$, $R^3 = (CH_3)_2C{=}CHCO$

3 $R^1 = H$, $R^2 = Ac$, $R^3 = (CH_3)_2C{=}CHCO$

4 $R^1 = R^2 = R^3 = H$

5 $R^1 = Me$, $R^2 = R^3 = H$

high-resolution mass and ^{13}C NMR spectrometry established a molecular formula of $C_{30}H_{48}O_8$. The IR spectrum showed the presence of a carboxylic acid (3500–2400 and 1700 cm^{-1}) and a methylenic double bond (1640 and 895 cm^{-1}). The ^1H NMR spectrum (Table I) exhibited resonances that could be assigned to six methyl groups, one singlet at δ 0.81, one doublet at δ 0.89 (J = 5.8 Hz), and four wide singlets at δ 1.68, 1.76, 1.87, and 2.13 that indicated vinylic methyls. Furthermore, observation of the δ 4–5 region, together with the number of oxygens present in the molecular formula, strongly suggested that spathulasin (1) contains a sugar moiety. Six ^{13}C NMR resonances at δ 114.02 (t), 115.86 (d), 121.96 (d), 136.15 (s), 147.80 (s), and 158.49 (s) pointed out the presence of three olefinic bonds in the molecule, a signal at δ 103.18 (d) was attributed to an anomeric sugar carbon, and two downfield signals at δ 166.47 (s) and 178.71 (s) were assigned to carboxylic groups.

Methylation of spathulasin (1, excess diazomethane in diethyl ether) yielded methyl ester **2** in high yield. The spectroscopic data of **2** confirm the aforementioned structural features.

Treatment of spathulasin (1) with excess acetic anhydride in pyridine at room temperature afforded diacetate **3** as shown by its ^1H NMR spectrum where two new methyl resonances at δ 2.00 and 2.05, corresponding to the esters formed, were observed.

Basic hydrolysis of spathulasin (1) gave **4**, which was characterized as its methyl ester **5**. From high-resolution mass spectrometry it was evident that the esterifying acid had a molecular formula of $C_5H_8O_2$. Comparison of the ^1H NMR spectra of **1** and **5** showed that the latter lacks the low-field methyl groups at δ 1.87 and 2.13 and the olefinic proton at δ 5.75. Also when the ^{13}C NMR spectra of these compounds were compared, the absence in **5** of five carbons at δ 20.56 (q), 27.60 (q), 115.86 (d), 158.49 (s), and 166.47 (s) was observed. These results indicate that the esterifying acid is 3,3-dimethylacrylic (senecioic) acid.[3]

Acid hydrolysis of methyl ester **5** produced cleavage of the sugar moiety to yield aglycon **6**. High-resolution mass

(1) Praeger, R. L. *An Account of the Sempervivum Group*; The Royal Horticultural Society: London, 1932.

(2) (a) Baker, A. J.; Eglinton, G.; González, A. G.; Hamilton, R. J.; Raphael, R. A. *J. Chem. Soc.* **1964**, 4705. (b) González, A. G.; Francisco, C. G.; Freire, R.; Hernández, R.; Salazar, J. A.; Suárez, E. *Phytochemistry* **1976**, *15*, 344.

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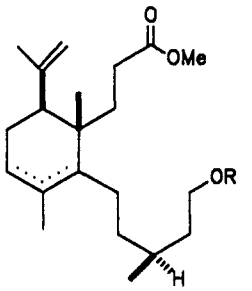
0022-3263/89/1954-5343$01.50/0

Table I. ^{1}H NMR Spectral Data and Assignments[a]

protons	1	2[b]	3	5	8	9	10
4-Me	1.76 (s)	1.76 (s)	1.77 (s)	1.77 (s)	1.77 (s)	1.75 (s)	1.77 (s)
7-H	5.35 (m)	5.35 (m)	5.36 (m)	5.39 (m)	5.39 (m)	5.45 (m)	5.43 (m)
8-Me	1.68 (s)	1.69 (s)	1.69 (s)	1.70 (s)	1.72 (s)	1.72 (s)	1.72 (s)
10-Me	0.81 (s)	0.84 (s)	0.84 (s)	0.83 (s)	0.87 (s)	0.86 (s)	0.86 (s)
13-Me	0.89 (d, J = 5.8)	0.92 (d, J = 6.5)	0.89 (d, J = 6.1)	0.89 (d, J = 5.9)	0.93 (d, J = 6.2)	0.95 (d, J = 6.3)	0.94 (d, J = 6.3)
15-H_2	3.49 (m), 3.88 (m)	3.52 (m), 3.95 (m)	3.46 (m), 3.90 (m)	3.49 (m), 3.89 (m)	3.44 (m), 4.03 (m)	3.51 (m), 4.08 (m)	3.52 (m), 4.09 (m)
18-H_2	4.77 (br s), 4.78 (br s)	4.77 (br s), 4.82 (br s)	4.78 (br s), 4.87 (br s)	4.77 (br s), 4.81 (br s)	4.77 (br s)	4.72 (br s)	4.72 (br s)
1′-H	4.23 (d, J = 6.8)	4.24 (d, J = 6.7)	4.40 (d, J = 6.8)	3.9[c]	4.25 (d, J = 7.9)	4.39 (d, J = 7.9)	4.40 (d, J = 7.9)
2′-H	3.66 (dd, J = 6.8, 9.0)	3.69 (dd, J = 6.8, 9.4)	5.19 (dd, J = 6.8, 9.4)	3.5[c]	4.92 (dd, J = 7.9, 10.1	5.41 (dd, J = 8.1, 10.7)	5.41 (dd, J = 7.8, 10.4)
3′-H	3.78 (dd, J = 3.4, 9.2)	3.81 (dd, J = 3.4, 9.3)	5.04 (dd, J = 3.4, 9.3)	3.5[c]	3.78 (dd, J = 3.6, 10.1)	5.20 (dd, J = 3.5, 10.6)	5.21 (dd, J = 3.5, 10.6)
4′-H	5.10 (m)	5.13 (m)	5.28 (m)	4.26 (m)	5.09 (m)	5.40 (m)	5.38 (m)
5′-H_2	3.58 (dd, J = 1.1, 13.2)	3.60 (dd, J = 1.6, 12.9)	3.65 (dd, J = 1.6, 12.9)	3.5[c]	3.53 (dd, J = 0.7, 13.9)	3.72 (dd, J = 1.1, 13.5)	3.72 (dd, J = 0.7, 13.4)
	3.98 (dd, J = 2.6, 13.2)	4.08 (dd, J = 3.2, 13.0)	4.07 (dd, J = 3.2, 13.0)	3.9[c]	4.10 (dd, J = 1.7, 13.6)	4.12 (dd, J = 1.9, 13.4)	4.12 (dd, J = 1.9, 8.3)
2″-H	5.75 (m)	5.78 (m)	5.77 (m)		5.78 (m)	5.78 (m)	5.79 (m)
3″-Me_2	1.87 (br s), 2.13 (br s)	1.89 (br s), 2.17 (br s)	1.91 (br s), 2.16 (br s)		1.92 (br s), 2.17 (br s)	1.89 (br s), 1.99 (br s)	1.90 (br s), 1.99 (br s)
OMe		3.65 (s)		3.66 (s)			
OAc			2.00 (s), 2.05 (s)				
Ar-H_4						7.54, 7.82 (AX, J = 8.5)	7.54, 7.81 (AX, J = 8.5)

[a] Assignments were aided by spin-decoupling and 2D COSY experiments. J values are reported in hertz, and chemical shift are given in δ units (downfield from TMS). [b] Recorded at 400 MHz. [c] Peak overlapping did not allow for an assignment of J values.

spectrometric analysis established $C_{21}H_{36}O_3$ as its molecular formula, which is consistent with the methyl ester derivative of a monocyclic diterpenoid acid with two double bonds and a hydroxyl group.



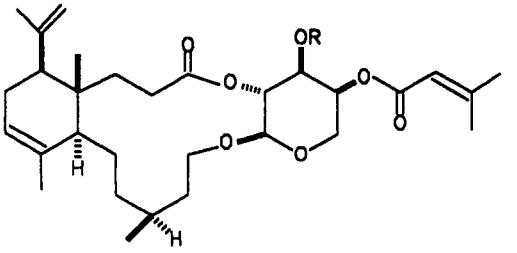
6 R = H

7 R = Ac

Some structural features can be inferred from the ^{1}H NMR spectrum of **6**, such as the presence of two double bonds (methylenic and trisubstituted), two vinylic methyl groups, and a primary hydroxyl group. Unfortunately, partial isomerization of the trisubstituted double bond to a tetrasubstituted position took place during acid hydrolysis, and the mixture could not be separated by chromatography on silica gel or silica gel–silver nitrate. Little connectivity could be established in the aglycon moiety by using ^{1}H NMR spin-decoupling and 2D COSY experiments. Since monocyclic diterpenes, apart from the cembrane group, are rare compounds, a secobicyclic diterpene was probably the most likely candidate to fulfill all spectroscopic data.[4]

The structure of the sugar portion of spathulasin (1), including the esterification site for 3,3-dimethylacrylic acid, was determined by ^{1}H NMR spin-decoupling and 2D COSY experiments summarized in Table I. The pentopyranose ring protons were assigned by decoupling experiments, and C-4′ was identified as the esterified carbon from the observation of the low-field shift of the ester methine proton (multiplet at δ 5.10). Furthermore, the observed coupling constants between the sugar protons (Table I) suggested that the pentose portion is 4′-*O*-(3,3-dimethylacryloyl)-α-arabinopyranose.[5] The ^{1}H NMR spectrum computer simulation of the sugar protons of compound **3** was in complete agreement with the original.[6]

Since the structure and stereochemistry of the asymmetric centers in the aglycone could not be assigned, X-ray crystallography was contemplated. Unfortunately, all attempts to prepare single crystals of the natural product **1** and derivatives **2** and **3** were unsuccessful. With this aim, spathulasin was treated with *p*-nitrobenzoyl chloride in dry pyridine to give compounds **8** and **9** in moderate yield.



8 R = H

9 R = p-$NO_2C_6H_4CO$

10 R = p-BrC_6H_4CO

(4) (a) Glasby, J. S. *Encyclopaedia of the Terpenoids*; Wiley: New York, 1982. (b) Devon, T. K. *Handbook of Naturally Occurring Compounds*; Vol. II; Academic: New York, 1972.

(5) Holland, C. V.; Horton, D.; Miller, M. J.; Bhacca, N. S. *J. Org. Chem.* **1967**, *32*, 3077.

(6) Calculated by using the program RACCOON by P. Schatz, University of Wisconsin, which is available through Project Seraphin, Eastern Michigan University, MI 48197.

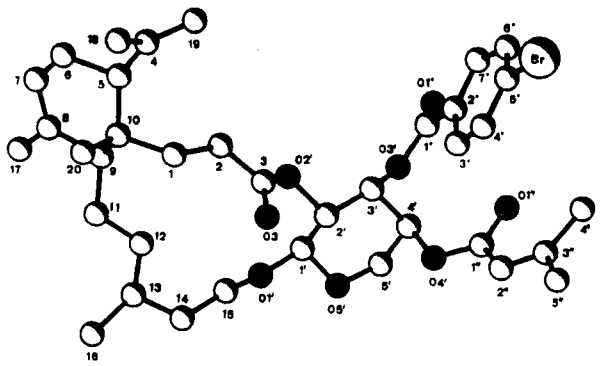


Figure 1. Computer-generated perspective drawing of lactone **10**. Hydrogen atoms are ommitted for clarity.

Compound **8** was analyzed by high-resolution mass spectrometry as $C_{30}H_{46}O_7$, which strongly suggested that an intramolecular lactonization of the acid group with one of the sugar hydroxyls had taken place. Study of the ^1H NMR spectrum of **8** allowed the location of the lactonization site at the C-2′ hydroxyl, as determined from the 1.26 ppm downfield shift observed for the corresponding methine proton (Table I). Furthermore, no downfield shift was observed for the proton at 3′. This interesting 14-membered lactone arose presumably from a mixed anhydride formed by the C-3 acid and the *p*-nitrobenzoyl chloride.[7]

Compound **9**, also obtained in the *p*-nitrobenzoylation reaction, was analyzed as $C_{37}H_{49}NO_{10}$ by high-resolution mass spectrometry, which corresponds to the 3′-*p*-nitrobenzoate derivative of **8**. As expected, a downfield shift was observed in comparing the chemical shift (5.20 ppm) of the 3′-methine proton in the ^1H NMR spectrum of **9** with that of **8** (3.78 ppm) (Table 1). Unfortunately, compound **9** gave only very small crystals, which were not suitable for X-ray analysis. Appropriate crystals were obtained from lactone **10** synthesized by reaction of **1** with *p*-bromobenzoyl chloride; its ^1H NMR spectrum was very similar (Table I) to that of **9**, and its high-resolution mass spectrum confirmed the structure of *p*-bromobenzoyl lactone, as shown.

A computer-generated perspective drawing of the final X-ray model of lactone **10** displaying the conformation of the 14-membered ring lactone is given in Figure 1. The absolute configuration corresponds to the selection of L-arabinose as the sugar component that is in the pyranose form and adopts a chair conformation. The terpene carbocyclic ring is in the half-chair conformation, with the isopropenyl group in an equatorial position.

The sugar was next isolated to determine its absolute configuration and hence the absolute configuration of all chiral centers in spathulasin (**1**). Hydrolysis of **4** was carried out with 1 N HCl/MeOH at 40–45 °C, the reaction mixture was passed through Amberlite exchange resin, and the aqueous methanolic solution was lyophilized. The residue was acetylated with acetic anhydride in pyridine and purified by Chromatotron chromatography to give methyl 2,3,4-tri-*O*-acetyl-β-L-arabinopyranoside and methyl 2,3,4-tri-*O*-acetyl-α-L-arabinopyranoside, identical with standard samples. A sample of L-arabinose was also treated with 1 N HCl, worked up, and acetylated under identical conditions to give the same methyl arabinopyranosides.[8]

(7) For other lactonizations of ω-hydroxy acids using acid chlorides see: (a) Inanaga, J.; Hirata, K.; Saeki, H.; Katsuki, T.; Yamaguchi, M. *Bull. Chem. Soc. Jpn.* **1979**, *52*, 1989. (b) Waanders, P. P.; Thijs, L.; Zwanenburg, B. *Tetrahedron Lett.* **1987**, *28*, 2409.

Since the relative stereochemistry of all centers is known by X-ray studies, and the sugar was L-arabinose, the absolute configuration of all chiral centers in spathulasin aglycone could be assigned as C_5 (*S*), C_9 (*S*), C_{10} (*S*), and C_{13} (*S*). Thus, the aglycone moiety of spathulasin represents a new 3,4-secolabdane diterpene.[9]

Experimental Section

General Methods. Melting points were determined with a Kofler hot-stage apparatus and are uncorrected. Optical rotation measurements were recorded at room temperature for solutions in $CHCl_3$ on Perkin-Elmer 141 and 142 polarimeters. IR spectra were recorded on Perkin-Elmer 257 and 681 spectrometers in $CHCl_3$ solutions. ^1H NMR spectra were recorded on Bruker WP 200 SY and I.E.F. (400 MHz) spectrometers, and ^{13}C NMR spectra on a Bruker WP 200 SY for solutions in $CDCl_3$ with Me_4Si as internal standard. Low-resolution mass spectra were determined with Hewlett Packard 5930 A and VG Micromass ZAB-2F spectrometers, and high-resolution mass spectra on a VG Micromass ZAB-2F spectrometer. Merck silica gels 60 and 0.063–0.2 mm were used for preparative thin-layer chromatography and column chromatography, respectively. Circular layers of 1 mm of Merck silica gel 60 PF 254 were used on a Harrison Chromatotron for centrifugally assisted chromatography. Commercial reagents and solvents were analytical grade or were purified by standard procedures[10] prior to use. *p*-Nitrobenzoyl and *p*-bromobenzoyl chlorides were prepared by the action of thionyl chloride upon the acid[11] and purified by fractional distillation.

Collection and Extraction. *A. Spathulatum* (35 kg), collected in February at Orotava, Tenerife, Canary Islands, was air-dried and extracted with EtOH in a Soxhlet apparatus. The EtOH extract was then filtered and evaporated under vacuum to give a residue (900 g) that was fractionated on a silica gel column. Fractions eluted with benzene/ethyl acetate, 1:1, were combined (4.5 g) and subsequently purified by chromatography (benzene/ethyl acetate, 4:1) to give spathulasin (**1**, 2.5 g).

Spathulasin (1): amorphous solid; $[\alpha]_D$ +43° (*c*, 0.452); IR 3500–2400 (br b), 1700, 1640, 1140, 1070, 895 cm^{-1}; MS, *m/z* (rel intensity) 536 (M^+, 1), 463 (1), 363 (2), 351 (3), 333 (2), 321 (13), 297 (5), 277 (3), 249 (6), 221 (8), 215 (100), 197 (12), 154 (20), 141 (30), 83 (19); exact mass calcd for $C_{30}H_{48}O_8$ 536.3348, found 536.3392; ^{13}C NMR 16.05 (q), 19.98 (q), 20.56 (q), 22.82 (q), 22.94 (q), 24.21 (t), 27.60 (q), 28.96 (t), 29.67 (t), 30.44 (d), 32.69 (t), 35.73 (t), 38.78 (t), 39.05 (s), 47.75 (d), 49.57 (d), 63.96 (t), 68.74 (t), 69.58 (d), 71.79 (d), 72.20 (d), 103.18 (d), 114.02 (t), 115.86 (d), 121.96 (d), 136.15 (s), 147.80 (s), 158.49 (s), 166.47 (s), 178.71 (s) ppm.

Methylation of Spathulasin (1). To a stirred solution of **1** (150 mg, 0.28 mmol) in ethyl ether (10 mL) was added excess of CH_2N_2 in diethyl ether. The organic layer was evaporated under vacuum, and the residue chromatographed on silica gel (benzene/ethyl acetate, 4:1) to give the methyl ester **2** (120 mg, 80%) as an amorphous solid: IR 3580, 3060, 1720, 1640, 1140, 1070, 895 cm^{-1}; MS, *m/z* (rel intensity) 550 (M^+, 2), 532 (1), 519 (2), 463 (3), 419 (1), 377 (4), 365 (11), 335 (28), 297 (15), 277 (6), 261 (6), 249 (13), 235 (12), 215 (100), 197 (50), 168 (56), 115 (73), 109 (66), 101 (52), 83 (98); exact mass calcd for $C_{31}H_{50}O_8$ 550.3505, found 550.3484; ^{13}C NMR 16.35 (q), 19.87 (q), 20.55 (q), 22.74 (q), 23.06 (q), 25.15 (t), 27.58 (q), 29.08 (t), 29.65 (t), 30.93 (d), 32.92 (t), 36.33 (t), 39.00 (s), 39.35 (t), 49.18 (d), 51.71 (q), 63.85 (t), 68.43 (t), 71.84 (d), 72.23 (d), 103.15 (d), 113.93 (t), 115.83 (d), 121.88 (d), 136.17 (s), 147.77 (s), 158.44 (s), 166.37 (s), 174.87 (s) ppm.

Acetylation of Spathulasin (1). Excess acetic anhydride was added to 100 mg (0.186 mmol) of **1** dissolved in 5 mL of dry pyridine with stirring. The reaction mixture was stirred overnight

(8) Buckingham, J. *Dictionary of Organic Compounds*, 5th ed.; Chapman and Hall: New York, 1982; Vol. 4, p 3734.

(9) A related dicarboxylic diterpene has been described, but its structure was claimed only on the basis of scant spectroscopic evidence. See: Bohlmann, F.; Zdero, C. *Chem. Ber.* **1976**, *109*, 1436.

(10) Perrin, D. D.; Armarego, W. L. P.; Perrin, D. R. *Purification of Laboratory Chemicals*, 2nd ed.; Pergamon: New York, 1982.

(11) Vogel, A. I. *Practical Organic Chemistry*, 3rd ed.; Longman: London, 1956; p 791.

at room temperature, quenched with ice, and extracted with CH_2Cl_2 (3 × 15 mL). The combined CH_2Cl_2 layers were then washed with 5% HCl (3 × 15 mL), water, and 5% aqueous $NaHCO_3$, dried (Na_2SO_4), filtered, and evaporated. Silica gel column chromatography of the organic residue (hexane/ethyl acetate, 4:1) gave the diacetate **3** (98 mg, 85%) as an amorphous solid: IR 3500–2400 (br b), 1740, 1705, 1640, 1240, 1140, 1055, 895 cm^{-1}; MS, *m/z* (rel intensity) 620 (M^+, 1), 560 (1), 547 (1), 333 (2), 321 (5), 299 (100), 270 (3), 217 (15), 211 (29), 170 (12), 157 (11), 155 (10), 139 (24), 83 (95); exact mass calcd for $C_{34}H_{52}O_{10}$ 620.3560, found 620.3563; ^{13}C NMR 16.34 (q), 19.54 (q), 20.50 (q), 20.88 (q), 20.92 (q), 22.72 (q), 22.99 (q), 25.11 (t), 27.60 (q), 28.90 (t), 29.61 (t), 30.58 (d), 32.73 (t), 36.23 (t), 38.98 (s), 39.42 (t), 47.71 (d), 49.23 (d), 63.41 (t), 66.89 (d), 68.01 (t), 69.61 (d), 70.60 (d), 101.20 (d), 114.02 (t), 115.74 (d), 121.87 (d), 136.17 (s), 147.74 (s), 158.47 (s), 165.73 (s), 169.69 (s), 170.44 (s), 178.89 (s) ppm.

Base Hydrolysis of Spathulasin (1). To 340 mg (0.634 mmol) of spathulasin (**1**) was added 20 mL of 5% KOH/MeOH, and the reaction mixture was stirred overnight. Upon workup, ice was added, and the solution was poured into 100 mL of H_2O, acidified, and extracted with CH_2Cl_2 (3 × 15 mL). The combined CH_2Cl_2 layers were dried (Na_2SO_4), filtered, and evaporated to yield the crude acid **4**, which was treated with excess of CH_2N_2 as described above for spathulasin. The residue was chromatographed on silica gel (ethyl acetate) to give the methyl ester **5** (175 mg, 59%) as an amorphous solid: IR 3570, 3440 (br b), 3060, 1725, 1165, 1080, 895 cm^{-1}; MS, *m/z* (rel intensity) 468 (M^+, 2), 437 (1), 395 (1), 381 (3), 377 (2), 365 (3), 335 (10), 305 (3), 249 (13), 235 (8), 221 (4), 168 (27), 119 (22), 109 (34), 83 (100); exact mass calcd for $C_{26}H_{44}O_7$ 468.3087, found 468.3092; ^{13}C NMR 16.21 (q), 19.72 (q), 22.64 (q), 22.91 (q), 24.97 (t), 28.89 (t), 29.47 (t), 30.76 (d), 32.65 (t), 36.20 (t), 38.79 (s), 39.24 (t), 47.43 (d), 48.95 (d), 51.62 (q), 65.30 (t), 67.88 (d), 68.19 (t), 71.37 (d), 72.91 (d), 102.87 (d), 113.81 (t), 121.75 (d), 135.94 (s), 147.59 (s), 174.76 (s) ppm.

Acid Hydrolysis of 5. Compound **5** (22 mg, 0.047 mmol) was dissolved in 1 N HCl/MeOH, and the reaction mixture was warmed to 40–45 °C for 15 h. The solution was cooled to room temperature, diluted with H_2O (30 mL), and extracted with CH_2Cl_2 (3 × 15 mL). The combined CH_2Cl_2 layers were then washed with 5% $NaHCO_3$, dried over Na_2SO_4, filtered, and evaporated. Chromatotron chromatography of the residue (hexane/ethyl acetate, 4:1) afforded an unseparable mixture of diterpenes **6** (13 mg, 82%; Δ^7:Δ^8 2:1; ^1H NMR) as an oil: IR 3580, 3060, 1720, 1625, 1170, 1040, 895 cm^{-1}; MS, *m/z* (rel intensity) 336 (M^+, 25), 321 (2), 308 (2), 305 (6), 287 (3), 263 (4), 249 (98), 235 (34), 225 (15), 221 (16), 207 (14), 195 (16), 189 (15), 175 (16), 163 (44), 133 (78), 121 (69), 119 (83), 109 (56), 107 (100), 93 (40), 81 (47), 69 (48), 55 (52), 41 (43); exact mass calcd for $C_{21}H_{36}O_3$ 336.2665, found 336.2675; ^1H NMR 0.80, 0.89 (s, s, 10-Me), 1.68, 1.60 (s, s, 8-Me), 1.76, 1.73 (s, s, 4-Me), 3.64 (m, 15-H_2), 3.66 (s, O-Me), 4.77, 4.83 and 4.62, 4.85 (4 × br s, 18-H_2), 5.34 (m, 7-H).

Treatment of Spathulasin (1) with *p*-Nitrobenzoyl Chloride. To a stirred solution of spathulasin (**1**, 60 mg, 0.112 mmol) in dry pyridine (2.5 mL) was added *p*-nitrobenzoyl chloride (60 mg, 0.323 mmol) and a catalytic amount of 4-(dimethylamino)pyridine (DMAP). The reaction mixture was stirred for 48 h at room temperature, quenched with ice, and extracted with CH_2Cl_2 (3 × 15 mL). The combined CH_2Cl_2 layers were then washed with 5% HCl (3 × 15 mL), water, and 5% aqueous $NaHCO_3$, dried (Na_2SO_4), filtered, and evaporated. Chromatotron chromatography of the residue (hexane/ethyl acetate, 19:1) gave the hydroxy lactone **8** (15 mg, 26%) and the *p*-nitrobenzoate lactone **9** (14 mg, 19%). Compound **8**: amorphous; IR 3560, 3060, 1715, 1640, 1520, 1150, 1080, 895 cm^{-1}; MS *m/z* (rel intensity) 518 (1), 439 (2), 351 (17), 333 (3), 321 (2), 315 (1), 269 (8), 215 (1), 197 (4), 185 (8), 137 (13), 119 (20), 107 (25), 95 (34), 83 (100); exact mass calcd for $C_{30}H_{46}O_7$ 518.3240, found 518.3306. Compound **9**: mp 197–199 °C (MeOH); $[\alpha]_D$ +111° (*c* 0.114); IR 1730, 1640, 1605, 1528, 1345, 1270, 1140, 1095, 895, 870 cm^{-1}; MS, *m/z* (rel intensity) 667 (M^+, 4), 652 (1), 637 (1), 500 (10), 470 (3), 418 (4), 385 (5), 364 (3), 333 (12), 150 (23), 137 (16), 120 (45), 107 (16), 95 (22), 83 (100); exact mass calcd for $C_{37}H_{49}NO_{10}$ 667.3352, found 667.3358.

Treatment of Spathulasin (1) with *p*-Bromobenzoyl Chloride. Compound **1** (400 mg, 0.746 mmol) in pyridine (10 mL) was treated with *p*-bromobenzoyl chloride (3 g, 13.6 mmol) and DMAP, as described previously, for 16 h. Chromatotron chromatography of the residue (benzene/ethyl acetate, 99:1) gave the *p*-bromobenzoate lactone **10** (87 mg, 17%) and a bis(*p*-bromobenzoate) derivative (79 mg), the structure of the latter remaining undetermined. Compound **10**: mp 167–169 °C (pentane); $[\alpha]_D$ +115° (*c* 0.22); IR 3060, 1715, 1640, 1585, 1265, 1140, 1095, 895 cm^{-1}; MS, *m/z* (rel intensity) 702/700 (M^+, bromine isotopes, 0.51/0.66), 603/601 (0.24/0.29), 535/533 (4.74/4.74), 453/451 (2.64/2.30), 399/397 (3.20/3.22), 333 (10), 202/200 (5.13/4.76), 185/183 (48.13/49.23), 137 (22), 119 (19), 107 (24), 95 (30), 83 (100); exact mass calcd for $C_{37}H_{49}BrO_8$ ($^{81}Br/^{79}Br$) 702.2585/700.2608, found 702.2606/700.2608; ^{13}C NMR 16.34 (q), 20.44 (q), 20.78 (q), 22.07 (q), 23.21 (q), 23.86 (t), 27.64 (q), 28.37 (t), 29.71 (t), 30.05 (d), 32.86 (t), 36.28 (t), 37.33 (t), 38.83 (s), 46.76 (d), 50.13 (d), 64.85 (t), 67.47 (d), 69.03 (t), 69.71 (d), 71.84 (d), 103.04 (d), 114.33 (t), 115.55 (d), 122.32 (d), 128.44 (s), 128.60 (s), 131.52 (dd), 131.88 (dd), 136.17 (s), 147.86 (s), 158.68 (s), 165.14 (s), 165.62 (s), 173.87 (s) ppm.

Hydrolysis of Spathulasin (1) To Give L-Arabinose. Compound **1** (200 mg, 0.37 mmol) was treated with 10 mL of 5% KOH/MeOH by the procedure outlined previously, and the residue treated with 1 N HCl/MeOH (3 mL) at 40–45 °C for 17 h. The reaction mixture was cooled to room temperature and passed through Amberlite IRA-400 (OH) exchange resin (Aldrich Chemical Co.) packing in a short column to remove the HCl. The resin was washed with MeOH (1 mL) and water (4 × 1 mL). The resulting aqueous methanolic solution was cooled in liquid nitrogen and lyophilized at room temperature for 24 h. Acetylation of the residue (131 mg), dissolved in dry pyridine, with excess of acetic anhydride as usual, gave after chromatotron chromatography (hexane/ethyl acetate, 19:1, 17:3) the diterpenes **7** [(61 mg, 43%) as an inseparable mixture (Δ^7:Δ^8 3:1; ^1H NMR)], methyl 2,3,4-tri-*O*-acetyl-β-L-arabinopyranoside (23 mg, 21%; $[\alpha]_D$ +168.4°, *c* 0.234; lit.[8] $[\alpha]_D$ +182°), and methyl 2,3,4-tri-*O*-acetyl-α-L-arabinopyranoside (7.5 mg, 8%; $[\alpha]_D$ +16.3°, *c* 0.08; lit.[8] $[\alpha]_D$ +19°). Compound **7**: amorphous; IR 3060, 1720, 1625, 1250, 1170, 895 cm^{-1}; MS, *m/z* (rel intensity) 378 (M^+, 41), 363 (3), 346 (9), 335 (5), 318 (9), 291 (59), 278 (8), 263 (21), 250 (19), 235 (31), 207 (21), 175 (28), 168 (55), 133 (62), 121 (90), 119 (91), 109 (95), 107 (100), 93 (88); exact mass calcd for $C_{23}H_{38}O_4$ 378.2770, found 378.2770; ^1H NMR 0.83, 0.91 (s, s, 10-Me), 1.70, 1.63 (s, s, 8-Me), 1.78, 1.76 (s, s, 4-Me), 2.04 (s, O-Ac), 3.66 (s, O-Me), 4.09 (m, 15-H_2), 4.77, 4.80 and 4.67, 4.90 (4 × br s, 18-H_2), 5.38 (m, 7-H) ppm. To check if the sugar could be isolated, conditions for reaction with 1 N HCl/MeOH were first worked with L-arabinose (Aldrich Chemical Co., 100 mg, 0.67 mmol), obtaining after acetylation and chromatotron chromatography (hexane/ethyl acetate, 17:3) methyl 2,3,4-tri-*O*-acetyl-β-L-arabinopyranoside (75 mg, 35%) and methyl 2,3,4-tri-*O*-acetyl-α-L-arabinopyranoside (27 mg, 14%) in all respects (R_f, $[\alpha]_D$, IR, and ^1H NMR) identical with the arabinopiranoside derivatives obtained from **1**.

Single-Crystal X-ray Structure Determination of the *p*-Bromobenzoate 10. All attempts to prepare single crystals from the native compound (**1**) failed. The *p*-nitrobenzoate **8** gave crystals that were too small, but *p*-bromobenzoate **10** led to bright rods that are found suitable for X-ray analysis. X-ray diffraction experiments were performed on a Philips PW 1100 automatic four-circle diffractometer operating with the Cu Kα radiation (λ = 1.5418 Å) monochromated by graphite. The orientation matrix of the crystal was calculated from the angular settings of 25 randomly distributed reflections found in the range $10° < \theta < 25°$ and refined by the least-squares procedure. No significant decomposition was found during the data collection, and no correction was applied. The reflections were scanned over a 1.2° angle width at a speed of 0.03° s^{-1}, and for each reflection, the background was deduced from two stationary measurements on both sides of the reflection. The intensities were reduced to *F* structural factors by standard Lorentz and polarization corrections and considered as observed above the 2σ background level. No absorption correction was applied. A crystal 0.6 × 0.4 × 0.3 mm in size was used. The cell parameters were as follows: a = 36.626 (6) Å; b = 16.656 (4) Å; c = 6.136 (3) Å; β = 99.6 (1)°. The space group was $P2_1$. The calculated volume of the cell, V = 3691.08 $Å^3$, can accommodate two independent molecules in the asymmetric unit (Z = 4) of composition $C_{37}H_{49}BrO_8$. A total of 9543

reflections were scanned up to $2\theta = 66°$ and led to 6223 unique structure factors ($R_{sym} = 5.6\%$) after data reduction. The corresponding normalized structure factors were calculated as usual (calcd scale factor = 0.38, mean $\langle U \rangle$ thermal factor = 0.032), and direct methods were applied. After many trials on phase sets corresponding to the highest figures of merit, we were able to develop a partial structure that gave upon F recycling procedure the whole set of atoms. The refinements of the structure were made with isotropic thermal factors for all the non-hydrogen atoms, in the first step, and then with anisotropic thermal factors with the Br and O atoms in a second step. The final R conventional factor converged to $R = 9.1\%$ for 2320 observed structural factors greater than 3σ. Due to the paucity of the data, no attempt was made to refine anisotropically the whole structure and carbon atoms were kept anisotropic.

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Supplementary Material Available: Tables of positional parameters, thermal parameters, interatomic distances, and interatomic angles for macrolactone 10 (5 pages); tables of observed and calculated structure factors (16 pages). Ordering information is given on any current masthead page.

Effect of *N*-Chloro Structure and 1-Substituent on σ-Substitution (Addition–Elimination) in Pyrroles[1]

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1-Methylpyrrole was reacted with a number of different types of *N*-chloro derivatives, and σ-substitution was observed only with *N*-chloroimides. It is proposed that there is a qualitative relationship between the pK_a of the *N*-chloro precursor and the reaction path. The reaction of 1-substituted pyrroles (CH_3, C_6H_5, $C_6H_5CH_2$, $CH_3C{=}O$, $(CH_3)_3C$, $CH_2{=}CHCH_2$, $(C_6H_5)_3C$) with *N*-chlorosuccinimide in $CHCl_3/NaHCO_3$ indicated that σ-substitution was very sensitive to electronic factors, e.g. no reaction was observed with 1-acetylpyrrole. It was not as sensitive to steric effects, and only with 1-tritylpyrrole was σ-substitution not observed. Only chlorination was observed with pyrrole itself.

Electrophilic substitution typically occurs by an S_E2 mechanism in aromatic[4] and heteroaromatic systems.[5] This is not the exclusive reaction path with electrophiles; addition[6,7] and substitution by σ-substitution[8] (addition–elimination[6,7]) are sometimes observed. Recently we reported that the reaction of 1-methylpyrrole with *N*-chloroacetanilide gave a product (ca. 20% yield) in which the acetanilide moiety had been incorporated into the pyrrole ring by σ-substitution (addition–elimination),[9] the first such example in pyrrole chemistry.[10,11] Addition–elimination reactions have been previously observed in benzene derivatives,[6-8] polyaromatic hydrocarbons,[6,7,8d] furans,[12-14] benzofurans,[15,16] and indoles.[17-19]

Table I. Reaction of *N*-Chloro Derivatives with 1-Methylpyrrole: Effect of *N*-Chloro Structure

N-chloro compound	pK_a[a]	% yield[c] base[d]	% yield[c] no base
N-chlorophthalimide (NCP)	8.30	81	66
N-chlorobenzotriazole	8.60	0[e]	0[e]
N-chlorosuccinimide (NCS)	9.70	74	33
N-chloromaleimide (NCM)	10.2[b]	79	64
N-chloroacetanilide[9]			20
N-chlorobenzimidazole	12.9	0[e]	0[e]
N-chlorobenzamide	14–15	NR[f,g]	0[e]
N-chloroacetamide	15.1	NR[g]	NR[g]
N-chlorourea		NR[g]	NR[g]
N-chloro-*N*,*N*′-dimethylurea		NR[g]	NR[g]

[a] Values taken (except where noted) from ref 29. [b] Calculated from data in ref 34. [c] Determined by ^{1}H NMR spectroscopy. [d] $NaHCO_3$. [e] Only chloropyrroles. [f] No reaction after 48 h. [g] Pyridine as base; no reaction in the absence of base under a N_2 atmosphere.

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