Rhabdastrellic Acid-A, a Novel Triterpenoid from the Marine Sponge *Rhabdastrella globostellata*

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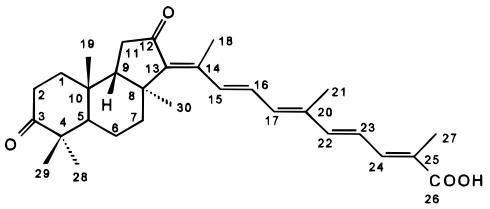
A new isomalabaricane triterpenoid, rhabdastrellic acid-A **(1)**, was isolated from the sponge *Rhabdastrella globostellata* (Carter) collected from the South China Sea near Hainan Island, People's Republic of China. Its structure was established on the basis of UV, IR, MS, ^{1}H-NMR, ^{13}C-NMR, and 2D NMR spectrometry. The relative and absolute stereochemistries were solved by NOESY and CD studies, respectively.

Marine sponges have proven to be a particularly fruitful source of unusual terpenoids.[1,2] Recently we examined the yellow sponge *Rhabdastrella globostellata* (Carter) and isolated a triterpenoid which is diastereomeric with a malabaricane triterpenoid reported from *Jaspis stellifera*.[3,4]

An EtOH extract of the dried sponge was partitioned between EtOAc and H_2O. Si gel chromatography of the EtOAc extract followed by Sephadex LH-20 column chromatography and recrystallization from a mixture of benzene and acetone afforded rhabdastrellic acid-A **(1)**.



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Compound **1** was obtained as a yellow crystals, $[\alpha]^{25}_D$ −61.6°; the molecular formula $C_{30}H_{40}O_4$ was established by HREIMS. Its IR spectrum revealed absorptions at 3427, 1710, and 1691 cm^{-1}, which were attributed to hydroxyl and conjugated ketone carbonyls, respectively. UV absorptions at 396nm (ϵ = 87 400) and 410 nm (ϵ = 85 200) suggested the presence of a conjugated polyene system. The ^{13}C-NMR spectrum of **1** revealed resonances at 219.19 (C-3, s), 207.15 (C-12, s), 173.36 (C-26, s), 146.77 (C-13, s), 144.37 (C-22, d), 141.75 (C-14, s), 140.60 (C-24, d), 138.55 (C-20, s), 135.58 (C-17, d), 134.71 (C-15, d), 131.90 (C-16, d), 126.55 (C-25, s), and 124.77 (C-23, d)ppm, indicating the presence of two ketone carbonyls, one carboxyl carbonyl and five carbon−carbon double bonds. Because the unsaturation number is 11, **1** must therefore be tricyclic, and the UV spectrum requires that all five carbon−carbon double bonds and at least one carbonyl be conjugated.

The ^{1}H-NMR spectrum of **1** revealed olefinic proton resonances at 6.76 (1H, d, J = 15 Hz), 7.06 (1H, dd, J = 11.8, 14.5 Hz), 6.44 (1H, d, J = 11.4 Hz), 6.659 (1H, d, J = 13.2 Hz), 6.676 (1H, dd, J = 13.2, 9.3 Hz), and 7.42 (1H, d, J = 9.3 Hz). Methyl signals at 2.37 (3H, s), 2.07 (3H, s) and 2.05 (3H, s) were typical of the ^{1}H-NMR spectrum of *E*-orientated carotenoids.[3] The other four methyl singlets at 1.46, 1.15, 1.08, and 0.88 ppm establish methyl groups connected with sp^3 carbons. Because the ^{13}C-NMR spectrum of **1** revealed only three singlets in the aliphatic region, it must contain a *gem*-dimethyl group. The protonated carbons of **1** were all assigned by heteronuclear multiple quantum coherence (HMQC) experiment.

A combination of ^{1}H−^{1}H DQFCOSY and ^{1}H-detected heteronuclear multiple-bond correlation (HMBC) experiments allowed the partial structure of the unsaturated branch of **1**. In the ^{1}H−^{1}H DQFCOSY spectrum, the protons at 6.76 and 6.44 ppm (labeled H-15 and H-17, respectively) revealed couplings to the proton at 7.06 ppm (H-16). In the HMBC spectrum the proton at 6.76 ppm (H-15) showed cross peaks with C-13, C-14, and C-18, and the proton of 7.06 ppm (H-16) showed coupling with C-14, so the connectivity from C-13 to C-18 could be determined. The ^{1}H−^{1}H DQFCOSY spectrum also revealed the couplings of protons at 6.659 ppm (H-22) and 7.42 ppm (H-24) with the proton at 6.676 ppm (H-23). HMBC experiment results revealed the coupling of H-24 with C-25, C-26, and C-27, and both H-17 and H-22 showed couplings with C-20.

The chemical shift of H_3-18 is downfield compared with normal methyls attached to carbon−carbon double bonds, while that of H-15 is that of a typical alkene proton. This indicates that H_3-18 must be in the deshielding region of carbonyl group and that the configuration of the C-13−C-14 double bond is thus *E*. If it were in the *Z*-configuration, the chemical shift of H-15 would be at a much lower field, because it would be in the deshielding region of the carbonyl group.[3,4]

Further evidence for the structure of **1** was based on an HMBC experiment. In the HMBC spectrum, both methyl proton signals at 1.15 and 1.08 ppm (CH_3-28 and CH_3-29, respectively) coupled with C-3, C-4, and C-5. The single methyl proton at 0.88 ppm (H-19) coupled with C-1, C-5, C-9, and C-10 (31.43, 45.55, 47.93, and 34.85 ppm, respectively). Another singlet methyl proton at 1.46 ppm (CH_3-30) showed connectivities to C-8, C-9, and C-13 (corresponding to 45.08, 47.93, and 146.77 ppm, respectively). The methyl proton (CH_3-18) in the chain coupled with C-13 indicating that the chain was

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attached to C-13. Connectivities between H-11 and C-12, and H-2 and C-3 were also revealed by the HMBC experiment.

A detailed analysis of the $^1H-^1H$ DQFCOSY spectrum showed connectivities of H-1 (2.19, 1.54 ppm) to H-2 (2.76, 2.41 ppm), H-5 (2.44 ppm) to H-6 (1.64, 1.54 ppm), H-6 to H-7 (2.21 ppm), H-9 (1.90 ppm) to H-7 (2.21 ppm), and H-9 (1.90 ppm) to H-11 (2.25 ppm).

The relative stereochemistry of **1** was elucidated by NOESY experiments. In the NOESY spectrum, the crosspeaks of H-18/H-16, H-16/H-21, H-21/H-23, and H-23/H-27 indicated an *E*-configuration of all the five carbon−carbon double bonds of the side chain. The cross peaks for H-5/H-28, H-5/H-30, H-9/H-19, and H-9/H-29 revealed 19-β-methyl, 30-α-methyl, and 9-β-H.

The absolute configuration was established by a CD study which showed an ϵ_{max} of 2.08 at 285 nm. The positive Cotton effect was in accordance with the analysis of the octant rule for ketones.

A similar compound to **1** was isolated by Ravi et al. from the sponge *Jaspis stellifera*,[3] and was later reassigned the isomalabaricane skeleton.[4] Compound **1** differs from Ravi's compound in having an *E* stereochemistry of the C-13−C-14 double bond.

Experimental Section

General Experimental Procedures. Melting point was recorded on a Reichert−Wien microscale melting point apparatus and temperature was uncorrected. Optical rotation was measured on a Perkin-Elmer 241MC polarimeter. IR spectra were recorded as KBr pellet on a Perkin-Elmer 983 spectrophotometer. UV spectra were recorded on a Perkin-Elmer Lambda 5 spectrophotometer. ^1H, ^{13}C, $^1H-^1H$ DQFCOSY, HMQC, HMBC, and NOESY NMR spectra were recorded on a Bruker AMX-600 MHz instrument in $CDCl_3$ with TMS as an internal standard. EIMS and HREIMS data were obtained on Quattro and Finnigan MAT 312 mass spectrometers, respectively. CD spectra were recorded on a JASCO J-500A instrument in absolute EtOH.

Animal Material. The dried yellow sponge *Rhabdastrella globostellata* (1.3 kg) was collected from the South China Sea near Hainan Island, People's Republic of China in 1992, and was identified by Dr. Rob Van Soest (Department of Coelenterates and Porifera, Institute for Systematics and Population Biology (Zoologisch Museum), University of Amsterdam). The voucher specimen (No. SS301) is deposited at the Guangzhou Institute of Chemistry, Chinese Academy of Sciences.

Extraction and Isolation. Chopped dried sponge (1.2 kg) was extracted with 95% ethanol. Concentration of the extract under reduced pressure gave 48.0 g of deep brown gum. The gum was evenly dispersed in 500 mL of distilled water and then was partitioned with EtOAc. The EtOAc-soluble fraction was concentrated under reduced pressure to give a crude residue (32.2 g), part of which (18.0 g) was chromatographed on a Si gel column with petroleum ether−EtOAc (100:0−70:30). The fraction of petroleum ether−EtOAc (80:20) was evaporated under reduced pressure to give a residue (2.6 g). The residue was rechromatographed on a Si gel column with petroleum ether−acetone (90:10 followed by 80:20). The eluate by petroleum ether−acetone (80:20) was rechromatographed on a Sephadex LH-20 column with $CHCl_3$−MeOH (30:70) and was then recrystallized from benzene−acetone to give pure **1** as yellow crystals (45 mg); mp 169−171 °C; $[\alpha]^{25}_D$ (c = 0.6, acetone) −61.6°; UV (acetone) $\lambda_{max}(\epsilon)$ 396 (87 400), 410 (85 200) nm; IR (KBr) ν_{max} 3427, 2939, 1710, 1691, 1579, 1422, 1391, 1289, 1183, 1160, 974, 803 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.42 (1H, d, J = 9.3 Hz), 7.06 (1H, dd, J = 11.8, 14.5 Hz), 6.76 (1H, d, J = 15.0 Hz), 6.676 (1H), 6.659 (1H), 6.44 (1H, d, J = 11.4 Hz), 2.76 (1H, m), 2.44 (1H, m), 2.37 (3H, s), 2.25 (2H, dd, J = 10.0, 12.1 Hz), 2.21 (2H, m), 2.07 (3H, s), 2.05 (3H, s), 1.90 (1H, dd, J = 10.0, 12.1 Hz), 1.64 (1H, m), 1.54 (2H, m), 1.46 (3H, s), 1.15 (3H, s), 1.08 (3H, s), 0.88 (3H, s) ppm; ^{13}CNMR ($CDCl_3$) δ 219.19 (C-3, s), 207.15 (C-12, s), 173.36 (C-26, s), 146.77 (C-13, s), 144.37 (C-22, d), 141.75 (C-14, s), 140.60 (C-24, d), 138.55 (C-20, s), 135.58 (C-17, d), 134.71 (C-15, d), 131.90 (C-16, d), 126.55 (C-25, s), 124.77 (C-23, d), 47.93 (C-9, d), 46.92 (C-4, s), 45.55 (C-5, d), 45.08 (C-8, s), 38.56 (C-7, t), 36.74 (C-11, t), 34.85 (C-10, s), 33.52 (C-2, t), 31.43 (C-1, t), 29.30 (C-28, q), 26.01 (C-30, q), 23.52 (C-18, q), 19.82 (C-6, t), 19.45 (C-29, q), 14.59 (C-19, q), 13.08 (C-27, q), 12.65 (C-21, q) ppm; EIMS (70 ev) m/z 464 M^+ (5), 431 (6), 365 (20), 241 (8), 159 (7), 119 (12), 91 (14), 69 (56), 55 (100), 43 (53); HREIMS (70 eV) m/z 464.2943 $[M]^+$, calcd for $C_{30}H_{40}O_4$, 464.2727.

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