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Tedarenes A and B: Structural and Stereochemical Analysis of Two New Strained Cyclic Diarylheptanoids from the Marine Sponge *Tedania ignis*

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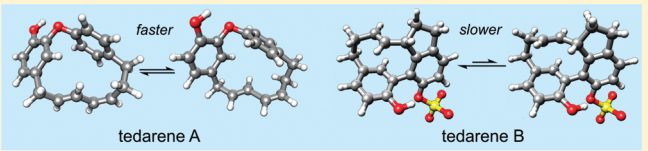
Supporting Information

ABSTRACT: Ring strain causes planar chirality in tedarenes A and B, two cyclic diarylheptanoids isolated from the marine sponge *Tedania ignis*. In both molecules, the chiral plane is an olefinic system, which is very rare among natural products. In tedarene A (**1**), interconversion is too fast to allow isolation of the enantiomeric atropisomers but still slow enough to cause coalescence of some ^{1}H and ^{13}C NMR signals at room temperature. In tedarene B (**2**), which also shows stable central and axial chirality, the two planar diastereomers are in slow equilibrium. Tedarene B is the smallest natural product with central, axial, and planar chirality in the same simple molecule. The identification of planar chirality as the difference between its conformational isomers allowed the use of theoretical prediction of the CD spectrum to determine the absolute configuration of the stereogenic carbon C-9 as well as of the biphenyl chiral axis.



INTRODUCTION

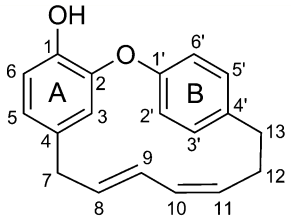
Diarylheptanoids are a class of natural products based on the 1,7-diphenylheptane skeleton that are mainly found in terrestrial plants. They are known to possess various biological activities, including leishmanicidal and antiprotozoal,[1] antitumor,[2] anti-inflammatory,[3] and inhibitory on nitric oxide production.[4,5] There exists a smaller number of cyclic diarylheptanoids that are formed from the corresponding linear type by phenolic oxidative coupling, either C−C coupling leading to *meta,meta*-bridged biaryls or C−O coupling leading to bridged diaryl ethers.[6]

We report here the isolation from the marine sponge *Tedania ignis* and the structural elucidation of two new cyclic diarylheptanoids, tedarenes A and B, which represent the first report of diarylheptanoids from marine sponges. Tedarenes A and B, which exist as equilibrium mixtures of atropisomers differing in their planar chirality, presented a number of stereochemical issues surprising for such small molecules. Tedarene A, but not tedarene B, showed an inhibitory effect against nitric oxide production.

RESULTS AND DISCUSSION

Tedania ignis was collected in the mangroves of Sweeting Cay (Bahamas) and extracted in sequence with MeOH and $CHCl_3$. Reversed-phase column chromatography, followed by repeated normal-phase and reversed-phase HPLC, gave pure tedarenes A (**1**) and B (**2**).

The high-resolution ESI mass spectrum of tedarene A (**1**) showed a pseudomolecular $[M + Na]^+$ ion peak at m/z 301.1205 (calcd 301.1199), determining the molecular formula



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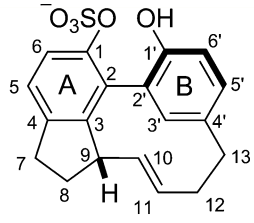

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$C_{19}H_{18}O_2$. The ^{1}H NMR spectrum (Figure 1a) showed signals of aromatic (δ 6.5−7.5) and olefinic (δ 5−6) protons. The COSY experiment led to the identification of a conjugated diene system (C-8/C-11) flanked by two methylene groups (C-7 and C-12). The configuration of the double bond at position 8 was determined as *E* ($J_{H\text{-}8/H\text{-}9}$ = 15.1 Hz) and that at position 10 as *Z* ($J_{H\text{-}10/H\text{-}11}$ = 11.0 Hz). ^{1}H signals at δ 6.52 (dd, J = 7.8 and 2.0 Hz, H-5), 6.64 (J = 7.8 Hz, H-6), and 5.37 (d, J = 2.0 Hz, H-3) suggested a 1,2,4-trisubstituted benzene, even though the chemical shift of H-3 is very low for an aromatic proton. The three nonprotonated carbon atoms of the benzene ring were identified through an HMBC spectrum. The HMBC and COSY experiments also connected the diene system with the benzene ring (correlation peaks of H_2-7 with C-3 and C-5 and long-range couplings of H_2-7 with H-3 and H-5).

While seven protons remained to be assigned, no more signals were evident in the ^{1}H NMR spectrum (Figure 1a). The presence of some very broad bands at δ 7.5−6.5 and δ 3.1−2.5 suggested the possibility of coalescent signals. Therefore, the

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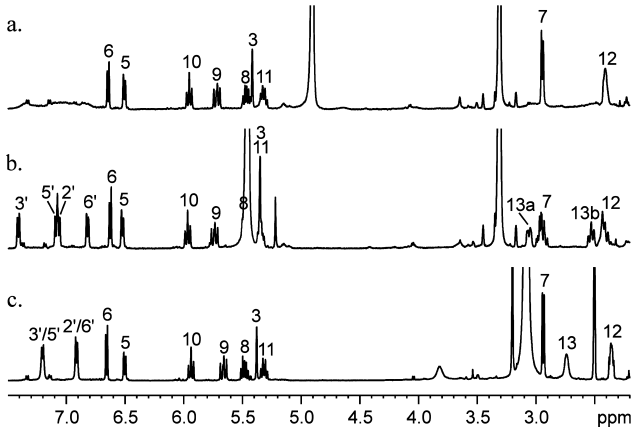

Figure 1. ^{1}H NMR spectra at 500 MHz of tedarene A (**1**) at (a) +20 °C (CD_3OD); (b) −40 °C (CD_3OD); (c) +80 °C (DMSO-d_6).

proton spectrum was measured at −40 °C to shift the conformational equilibrium to the slow exchange regime. At −40 °C, the broad humps turned into sharp ^{1}H signals (Figure 1b). A new set of 1D and 2D NMR experiments was acquired (Table 1 and Figure 2), showing the presence of a further *para*-disubstituted benzene ring (ring B) linked to the diene system through a further methylene group (CH_2-13).

The chemical shift of the three aromatic carbons C-1, C-2, and C-1′ suggested them to be linked to oxygen. By consideration of molecular formula, two of them should be involved in a diphenyl ether functional group, and one in a phenol functional group. The correlation peak of H-3 with H-6′ observed in the ROESY spectrum is geometrically incompatible with a C-1/C-1′ ether bridge and therefore located the ether bridge between C-2 and C-1′.

The ^{1}H NMR spectrum shows that, at −40 °C (Figure 1b), the three pairs of methylene protons are diasterotopic, and the

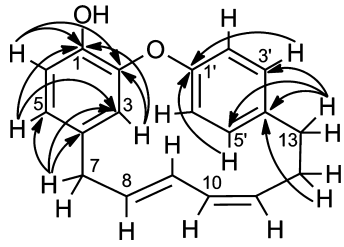

Figure 2. Most significant HMBC correlations detected for tedarene A (**1**).

apparently symmetric H-2′/H-6′ and H-3′/H-5′ pairs are also nonequivalent, pointing to a chiral structure. Analysis of the structure of tedarene A reveals the presence of planar chirality: the conjugated double bond system cannot reside in the plane of symmetry of the molecule because of ring strain. The molecule can therefore exist in two enantiomeric conformational isomers, which can interconvert by "flipping" the conjugated double bond. A rough estimation of the energy barrier between the conformational enantiomers based on Eyring equation[7] (coalescence temperature of 25 ± 5 °C for H-3′ and H-5′, $\Delta\nu$ = 150 Hz at 500 MHz) gave a value of $\Delta G^{\ddagger}$ = 14.0 ± 0.3 kcal/mol.

This idea was confirmed by the ROESY spectrum recorded at −40 °C, showing clear exchange peaks between H-3′ and H-5′ and between H-2′ and H-6′. In addition, in the ^{1}H NMR spectrum recorded at +80 °C the molecule is in the fast exchange regime and all the pairs of methylene protons are equivalent (Figure 1c).

To investigate the conformational behavior of **1**, the molecule was submitted to a molecular dynamics (MD) simulation for 100 ns at 400 K (the conformational equilibrium was too slow at 300 K), and 2000 structures were generate by extracting geometries from the MD trajectory every 50 ps. A

Table 1. ^{1}H and ^{13}C NMR Spectroscopic Data (500 MHz) for Tedarene A (1)

			−23 °C (250 K), CD_3OD				80 °C, DMSO-d_6
position			δ_H [mult, J (Hz)]	δ_C	HMBC[a]	ROESY	δ_H [mult, J (Hz)]
1	C			144.5			
2	C			151.6			
3	CH		5.37 (m)	117.7	1, 2, 4, 5, 7	9, 7b, 2′, 5′, 6′	5.38 (d, 1.9)
4	C			131.3			
5	CH		6.52 (dd, 7.8, 1.2)	121.5	1, 3, 7	7a	6.50 (dd, 7.9, 1.9)
6	CH		6.64 (d, 7.8)	116.1	1, 2, 4		6.65 (d, 7.9)
7	CH_2	a	2.97 (dd, 13.7. 6.3)	36.9	3, 4, 5, 8, 9	5, 11	2.94 (d, 7.9)
		b	2.93 (dd, 13.7, 9.7)			3, 8, 9	
8	CH		5.46 (m)	133.8	7, 10	7b, 10	5.48 (15.1, 7.9, 7.9)
9	CH		5.71 (dd, 15.1, 11.0)	129.6	7, 10	3, 7b, 12	5.66 (dd, 15.1, 11.0)
10	CH		5.95 (t, 11.0)	131.8	8, 9, 12	8, 11	5.94 (dd, 11.0, 11.0)
11	CH		5.34 (ddd, 11.0, 8.6, 8.6)	129.3		7a, 10, 12, 13b	5.31 (ddd, 11.0, 8.6, 8.6)
12	CH_2		2.41 (m)	33.0	10, 11, 13, 4′	9, 11, 13a, 5′	2.35 (m)
13	CH_2	a	3.06 (br dd, 12.0, 4.7)	35.9		12, 13b, 3′	2.74 (br s)
		b	2.52 (ddd, 12.0, 12.0, 2.1)		11, 12, 3′, 4′, 5′	11, 13a, 5′	
1′	C			156.3			
2′	CH		7.06 (dd, 8.3, 1.9)	125.5	4′, 6′	3	6.91 (d, 7.7)
3′	CH		7.38 (dd 8.3, 1.3)	131.4	13, 1′, 5′	13a	7.20 (d, 7.7)
4′	C			141.0			
5′	CH		7.08 (dd, 8.3, 1.3)	133.3	13, 1′, 3′	3, 12, 13b	7.20 (d 7.7)
6′	CH		6.81 (dd, 8.3, 1.9)	123.7	1′, 2′, 4′	3	6.91 (d, 7.7)

[a]HMBC correlations are from proton(s) stated to indicated carbon.

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flexible orientation of ring A was observed (the dihedral angle C3−C4−C7−C8 was found anywhere between −110° and +110°). More interestingly, flipping of the conjugated double bond system, and therefore inversion of planar chirality, occurred 36 times during the 100 ns simulation (Figure 3). When minimized, all the MD structures converged onto four pairs of enantiomeric conformers, all very similar. The lowest energy conformers are those depicted in Figure 3; the others (Figure S1 in the Supporting Information) differed only in minor details, i.e., the orientation of the OH bond (+1.7 kcal/mol), the orientation of ring A (+3.0 kcal/mol), or both (+4.7 kcal/mol). It must be noted that in each of these conformations H-3 lies in the shielding cone of the aromatic ring B, and this accounts for its unusually low chemical shift.

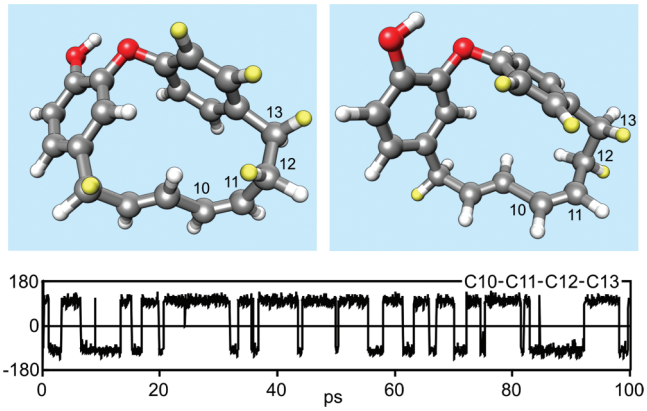

Figure 3. Two enantiomeric minimum energy conformers of tedarene A (**1**) and a plot of the C10−C11−C12−C13 torsion angle as a function of time during the MD simulation (400 K, 100 ns). A change in the sign of this torson angle indicated a change in the planar chirality of tedarene A. The two enantiomeric conformations are oriented so that the mirror plane is the paper plane, i.e., a bond that points upward in one conformation will point downward in the other. One of each pair of protons involved in chemical exchange is marked in yellow.

The negative-ion high-resolution ESI mass spectrum of tedarene B (**2**) showed an M^- molecular ion peak at m/z 357.0796 corresponding to $C_{19}H_{17}O_5S$ (calcd m/z 357.0802). Examination of the protons spectrum showed the presence of two conformers in slow exchange, with a 4:1 ratio as estimated by integration of NMR peaks. We did succeed in isolating the major isomer, but after 24 h the ^{1}H NMR spectrum showed again the equilibrium mixture. Therefore, we decided to study the conformers together.

Signals of the major conformer (Table 2) were analyzed first. Analysis of ^{1}H and COSY spectra allowed the identification of one 1,2,4-trisubstituted (oxygenated at C-1′, δ 154.2) and one 1,2,3,4-tetrasubstituted (oxygenated at C-1, δ 149.9) benzene ring (Table 2). In addition to the signals of the two aromatic rings, the NMR spectra also showed signals of an *E* double bond (C-10, δ 140.6 and C-11, δ 126.0; H-10, δ 3.86 and H-11, δ 5.34; $J_{H10/H11}$ = 16.6 Hz). The former proton resonated at an exceptionally shielded chemical shift for an olefinic proton. Combined analysis of COSY and HSQC spectra defined the aliphatic chain C-7/C-13 as CH_2−CH_2−CH−CH═CH−CH_2−CH_2. HMBC data (Figure 4) connected C-13 to C-4′, while the other aromatic ring was connected to the chain in two points (C-9 with C-3 and C-7 with C-4), thus defining a further five-membered ring. Finally, the two rings were directly linked

Table 2. ^{1}H and ^{13}C NMR Spectroscopic Data (700 MHz, CD_3OD) for Tedarene B (Major Conformer, *SSS*-2)[a]

position[b]			δ_H [mult, *J* (Hz)]	δ_C	HMBC[c]	ROESY
1	C			149.8		
2	C			130.8		
3	C			150.4		
4	C			142.5		
5	CH		7.20 (br d, 8.3)	125.4	1, 3, 6, 7	
6	CH		7.46 (br d, 8.3)	120.5	1, 2, 4	
7	CH_2	α	2.86 (br dd, 15.2, 7.8)[d]	31.8	3, 4, 5, 8, 9	8α, 8β, 5′
		β	2.78 (br ddd, 15.2, 11.9, 6.4)		1, 3, 4, 5, 8	8α, 8β, 9
8	CH_2	α	2.17 (dddd, 13.5, 11.9, 11.4, 7.8)	32.9	7, 9, 10	7α, 7β, 11
		β	1.98 (ddd, 11.4, 6.4, 5.8)[d]		3, 4, 7, 9	7α, 7β, 9
9	CH	β	3.35 (br ddd, 13.5, 7.2, 5.8)	48.3	2, 3, 8, 10, 11	7β, 8β, 10
10	CH		3.86 (dd, 16.6, 7.2)	140.2	8, 9, 11, 12	9, 12β, 3′
11	CH		5.34 (dddd, 16.6, 9.3, 5.6, 1.0)	126.0	9, 12	8α, 12α, 3′
12	CH_2	α	2.48 (dddd, 12.1, 5.6, 4.1, 2.7)	37.0	10, 11, 13, 4′	12β, 13β, 11
		β	1.49 (ddddd, 12.5, 12.1, 9.4, 4.7, 1.0)		10, 11, 13, 4′	10, 12α, 13β
13	CH_2	α	2.35 (ddd, 12.5, 12.5, 4.1)	38.0	11, 12, 3′, 4′, 5′	13β, 3′
		β	2.88 (ddd, 12.5, 4.7, 2.7)		11, 12, 3′, 4′, 5′	12α, 12β, 13α
1′	C			154.1		
2′	C			120.0		
3′	CH		6.08 (d, 2.1)	145.1	2, 13, 1′, 5′	10, 11, 13α
4′	C			131.9		
5′	CH		6.96 (dd, 8.1, 2.1)	129.2	13, 1′, 3′	7α
6′	CH		6.80 (d, 8.1)	118.3	1′, 2′, 4′	

[a]For NMR data of the minor conformer (*SSR*-**2**) see Table S1 in the Supporting Information. [b]Assignment of methylene protons is stereospecific. Protons on the same side as H-9 are denoted as β; protons on the opposite side are denoted as α. [c]HMBC correlations are from proton(s) stated to indicated carbon. [d]The coupling constant between H-7α and H-8β is close to 0, suggesting a dihedral angle close to 90° between the two protons. This was confirmed by molecular modeling.

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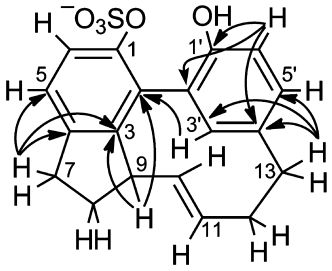

Figure 4. Most significant HMBC correlations detected for tedarene B (**2**).

through C-2 and C-2′, as shown by the HMBC interaction of H-3′ (δ 6.08) with C-2 (δ 130.8). A sulfate group, required by the molecular formula, was located at C-1 because of the deshielded chemical shift of the *ortho* proton H-6 (δ 7.46), while a hydroxyl group must be linked at C-1′, *ortho* to the shielded H-6′ (δ 6.80).

The structure of minor conformer was determined in a similar way, and all the ^{1}H and ^{13}C NMR signals were assigned (Table S1 in the Supporting Information). The planar structure (including the configuration of the double bond) was the same, but in this case it was H-11, and not H-10, that resonated at an unusually low chemical shift (δ 3.96).

Analysis of the structure of tedarene B reveals the presence of three chiral elements: one stereogenic carbon (C-9), one axially chiral biphenyl unit, and one planarly chiral double bond, leading to four potential atropisomers for each of the two configurational stereoisomers at C-9.[8] The four possible diastereomeric atropisomers of the configurational stereoisomer 9-*S*, namely (9*S*,2S_a,10S_p), (9*S*,2S_a,10R_p), (9*S*,2R_a,10S_p), and (9*S*,2R_a,10R_p) [referred to below as *SSS*-**2**, *SSR*-**2**, *SRS*-**2**, and *SRR*-**2**, respectively, Figure 5] were modeled and studied by MD simulations (100 ns). No conversion between conformational stereoisomers took place in simulations at up to 1200 K. At 1500 K, double bond flipping interconverting *SSS*-**2** and *SSR*-**2** was observed several times. On the other hand, *SRR*-**2** and *SRS*-**2** both inverted their axial chirality during the simulation (*SRS*-**2** also inverted its planar chirality at the very beginning of the simulation) and never returned to their original 2R_a conformation. These data clearly suggest that the conformational equilibrium of **2** involves a change in planar chirality (flipping of the double bond) and in particular exchange between *SSS*-**2** and *SSR*-**2**.

This computational analysis was confirmed experimentally by the ROESY correlation peaks observed for the two conformers of **2** and shown in Figure 6. For the major conformer, if the configuration at C-9 is assumed to be *S*, the correlation peaks H-9/H-10 and H-8α/H-11 defined the planar chirality of the double bond as 10S_p, while the correlation peak of H-11/H-3′ indicated the 2R_a axial chirality. Therefore, the major conformer was assigned to *SSS*-**2** (or its enantiomer). Likewise, the correlation peaks H-9/H-11, H-8α/H-10, and H-10/H-3′ assigned the minor conformer to *SSR*-**2** (or its enantiomer). In addition, H-10 in *SSS*-**2** and H-11 in *SSR*-**2** are in the shielding cone of the aromatic ring B, and this nicely explains the very shielded chemical shift of these two protons.

The detailed knowledge of the conformational behavior of tedarene B (**2**) was also the key to assign its absolute configuration. Compound **2** is optically active ($[\alpha]^{25}_{D}$ +33) and shows a clear CD spectrum (Figure 7a). A theoretical prediction of the CD spectra of *SSS*-**2** and *SSR*-**2** was obtained

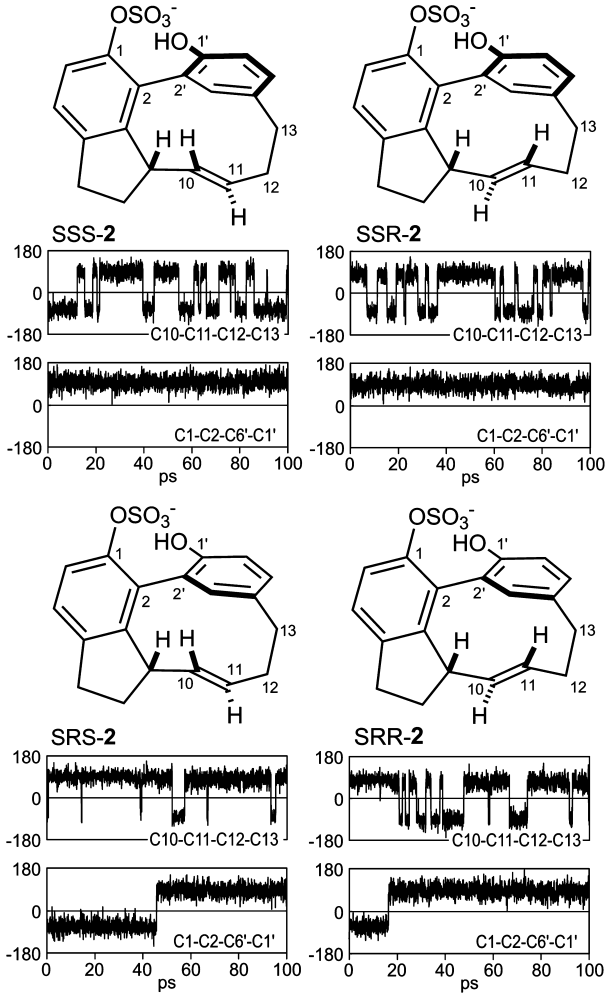

Figure 5. Four conformational stereoisomers of (9*S*)-tedarene B (**2**) and their conformational behavior during MD simulations (1500 K, 100 ns). The C10−C11−C12−C13 torsion angle is related to planar chirality; it is positive for the 10R_p, and negative for the 10S_p conformation. The C1−C2−C2′−C1′ torsion angle is related to axial chirality; it is positive for the 2S_a and negative for the 2R_a conformation.

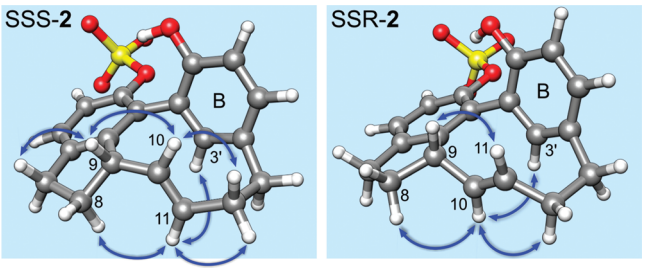

Figure 6. Diagnostic ROESY correlations detected (a) for the major conformer *SSS*-**2** and (b) for the minor conformer *SSR*-**2** of tedarene B.

by time-dependent density functional theory (Gaussian 03,[9] B3LYP functional, 6-31G* basis set), and the mean of the two spectra was calculated, weighed by the relative abundance of the two conformers as determined by ^{1}H NMR (Figure 7b). The predicted spectrum has the same general aspect of the experimental spectrum, with the same sign for each of the

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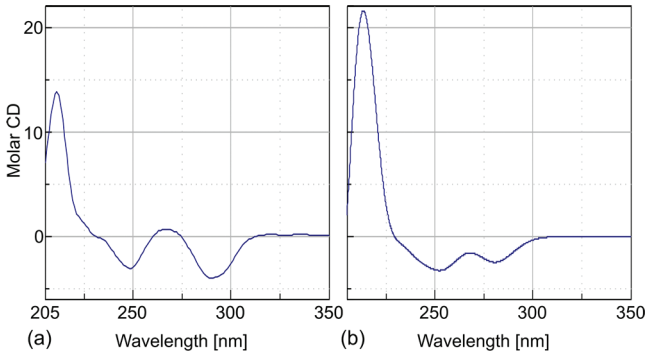

Figure 7. Experimental (a) and theoretical (b) CD spectra of tedarene B (**2**).

three bands. Therefore, the absolute configuration of tedarene B (**2**) is (9*S*,2S_a).

Tedarenes A and B were tested for their inhibitory activity against LPS-induced NO release from murine monocyte/macrophages J774 cells. The cells were incubated with LPS (1 μg/mL) and different concentrations of tedarenes. A significant inhibition of NO_2^- production was observed for tedarene A (**1**) at 10 and 30 μM, while tedarene B (**2**) was found to be inactive (Figure 8).

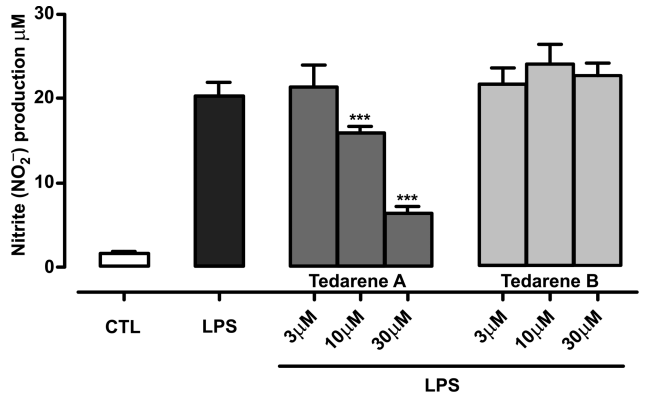

Figure 8. Effect of different concentrations of tedarene A (**1**) and tedarene B (**2**) on the production of NO_2^- by J774 macrophages stimulated with LPS (1 μg/mL). Each point represents the mean ± SEM of three separate experiments run in triplicate.

CONCLUSION

The structure of the two diarylheptanoids, tedarenes A and B, isolated from the marine sponge *Tedania ignis* was fully elucidated by a combination of spectroscopic and computational methods.

Structure elucidation of tedarene A is a nice example of deductive work that starts from a quite common experience, some missing NMR signals. While slow interconversion between conformational isomers and coalescence of NMR signals are very well studied, one does not expect to encounter this phenomenon in the study of a simple natural product. This is particularly true because only some protons and carbons are involved in the chemical exchange and therefore experience coalescence, while the others give sharp and well-resolved signals at any temperature.

Planar chirality has been previously described for some ether-bridged diarylheptanoids,[10] including galeon.[11,12] In these compounds, however, the atropisomers differ in the orientation of ring B, while in tedarene A (**1**) ring B is symmetrical, and it is the orientation of the conjugate double bond system that is different (Figure 9). As a matter of fact, an olefinic chiral plane

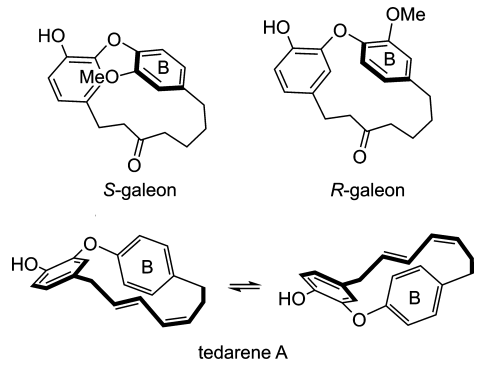

Figure 9. Planar chirality in galeon and tedarene A (**1**).

(like that in *trans*-cyclooctene) is very rare among natural products. The energy barrier between the two atropisomers of **1** is also much lower than for galeon, and they can interconvert at room temperature. ^{1}H and ^{13}C NMR spectra of **1** are strongly affected by this phenomenon, because the interconversion rate happens to be in the coalescence region at room temperature, causing some signals to become so broad as to be hardly detectable.

Chirality of macrocyclic biphenyls, such as tedarene B, may be very complex.[13] For example, isoplagiochin C was shown to occur in the liverwort *Plagiochila deflexa* as an 85:15 mixture of stable enantiomeric forms, each of which is a mixture of four conformational diastereomers in dynamic equilibrium.[14] The picture is much simpler for tedarene B (**2**), with only two conformational diastereomers in slow equilibrium. However, tedarene B (**2**) also contains a stereogenic carbon and is therefore characterized by the rare co-occurrence of central, axial, and planar chirality in the same simple molecule. To the best of our knowledge, **2** is the smallest natural product to possess this feature. The detailed study of dynamic chirality of **2** and the identification of planar chirality as the difference between its conformational isomers allowed us to use theoretical prediction of the CD spectrum to determine the configuration of the stereogenic carbon C-9.

EXPERIMENTAL METHODS

General Experimental Procedures. High-resolution ESI-MS spectra were performed on a LT-Orbitrap mass spectrometer. The spectra were recorded by infusion into the ESI source using MeOH as the solvent. Optical rotations were measured at 589 nm using a 10-cm microcell. CD spectra were recorded using a 1-cm cell. NMR spectra were determined at 700 and 500 MHz; chemical shifts were referenced to the residual solvent signal (CD_3OD: δ_H 3.31, δ_C 49.0; DMSO-d_6: δ_H 2.50). For an accurate measurement of the coupling constants, the one-dimensional ^{1}H NMR spectra were transformed at 64K points (digital resolution: 0.09 Hz). Through-space ^{1}H connectivities were evidenced using a ROESY experiment with a mixing time of 450 ms. The HSQC spectra were optimized for $^1J_{CH}$ = 142 Hz, and the HMBC experiments for $^{2,3}J_{CH}$ = 8.3 Hz.

Collection, Extraction, And Isolation. Specimens of *Tedania ignis* were collected in the mangroves of Sweeting Cay (Grand Bahama Island) during the 2007 Pawlik expedition. They were frozen immediately after collection and kept frozen until extraction. The

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sponge (121 g of dry weight after extraction) was cut into small pieces, and the extraction was carried out using MeOH (5 × 2 L), followed by $CHCl_3$ (2 × 2 L), as solvent. The MeOH extracts were partitioned between H_2O and BuOH, and the BuOH layer was combined with the $CHCl_3$ extract and concentrated in vacuo. The organic extract (29.5 g) was chromatographed on a column packed with RP-18 silica gel. A fraction eluted with MeOH/H_2O (8:2) (271 mg) was subjected to reversed-phase HPLC [RP-18, MeOH/H_2O (8:2)], thus affording two fractions containing, respectively, compound **1** and **2**. Pure tedarene A (**1**, 2.3 mg) was obtained by normal-phase HPLC on SiO_2, using hexane−2-propanol 95:5 as eluent. Compound **2** was purified by reversed-phase HPLC on an RP-18 column, using MeOH/H_2O (1:1) as solvent. Final HPLC purification was carried out using a pentafluorophenyl (PFP) column (100 × 4.6 mm, 2.6 μm) [MeOH/H_2O (1:1), flow 0.6 mL/min], giving a fraction composed of pure compound **2** (1.1 mg). The ^{1}H NMR spectrum of the fraction, recorded immediately after HPLC separation, showed it to be composed of *SSS*-**2**, with only trace amounts of *SSR*-**2**. However, after 24 h the ^{1}H NMR spectrum displayed a 4:1 equilibrium mixture of *SSS*-**2** and *SSR*-**2**.

Tedarene A (**1**): colorless amorphous solid; ^{1}H and ^{13}C NMR data, Table 1; UV (MeOH) $\lambda_{max}(\varepsilon)$ 288 (10200, shoulder), 257 nm (26600, shoulder); high-resolution ESI-MS (positive ion mode, MeOH) *m*/*z* 301.1205 ([M + Na]$^+$, $C_{19}H_{18}O_2Na$ gives 301.1199.

Tedarene B (**2**): colorless amorphous solid; $[\alpha]^{20}_D$ +33 (*c* 0.1, MeOH); ^{1}H and ^{13}C NMR data major conformer (*SSS*-**2**), Table 2; minor conformer (*SSR*-**2**), Table S1 (Supporting Information); UV (MeOH) $\lambda_{max}(\varepsilon)$ 281 nm (5500); CD (MeOH) $\lambda_{max}(\Delta\varepsilon)$ 290 (−4.0), 248 (−3.1), 211 nm (+13.8); high-resolution ESI-MS (negative ion mode, MeOH) *m*/*z* 357.0796 (M$^-$, $C_{19}H_{17}O_5S$ gives 357.0802).

Molecular Dynamics Simulations. MD calculations were performed using the CVFF force field[15] and the INSIGHT II/Discover package.[16] All of the simulations were performed in vacuo. For tedarene A (**1**), a first MD simulation was performed at 300 K for 100 ns, and the coordinates produced by the simulation were saved every 50 ps, giving 2000 structures. No change was observed in the planar chirality of the molecule during the simulation. This was not surprising because NMR data suggested an average time between inversions of chirality much longer than 100 ns at room temperature. Therefore, the simulation was repeated at 400 K, and this time inversion of chirality was observed several times. Each structure saved from MD was then minimized, and they all converged into four pairs of enantiomeric conformations.

As for tedarene B (**2**), after some trials we chose to perform MD simulations at 1500 K because at lower temperature the conformational changes were too slow. Four 100-ns simulation were performed using the four atropisomers of 9*S*-**2** (*SSS*-**2**, *SSR*-**2**, *SRS*-**2** and *SRR*-**2**, see the main paper) as starting structures. The coordinates were saved every 50 ps, leading to four sets of 2000 structures. At this temperature, it was necessary to restrain the double bond at position 10 to *trans* during the simulation because if tended to isomerize to *cis* in a free simulation. The restraint was removed before the subsequent minimization, and the lowest energy conformations obtained for *SSS*-**2** and *SSR*-**2** were used as starting structures for the quantum mechanical calculations.

Quantum Mechanical Prediction of the CD Spectrum. The lowest energy structures of the major and minor conformer of tedarene B (*SSS*-**2** and *SSR*-**2**, respectively) obtained from molecular mechanics were optimized using the Gaussian03W package[8] at the B3LYP/6-31G(d) level. The theoretical CD spectrum was calculated on the optimized geometries of the two conformers using the same B3LYP functional, the 6-31G(d,p) basis set, and the IEF-PCM model for the solvent (MeOH). The CD curves were calculated from the Gaussian output using the program SpecDis 1.45.[17] The the exponential half-width of the CD bands, σ, was selected as 0.25 eV by visual fitting with the experimental CD spectrum. Finally, the weighted mean of the theoretical CD curves was calculated based on the relative amounts of the conformers, as determined by integration of the signals in the ^{1}H NRM spectrum of **2**.

Cell Culture. The murine monocyte/macrophages J774 cells were grown in Dulbecco's modified Eagle's medium and cultured at 37 °C in humidified 5% CO_2/95% air. The cells were plated in 24 well culture plates at a density of 2.5 × 10^6 cells/mL/well and allowed to adhere for 2 h. Thereafter, the medium was replaced with fresh medium and cells were activated by lipopolysaccharide (LPS 1 μg/mL) from *E. coli* for 24 h in the presence or absence of different concentrations of test compounds. The culture medium was then removed and centrifuged, and the supernatant was used for the determination of nitrite (NO_2^-) production. Cell viability (>95%) was determined with the MTT assay.[18]

NO_2^- Assay. NO_2^- levels in culture media from J774 macrophages were measured 24 h after LPS with the Griess reaction as previously described.[19] Results are expressed as nmol/mL of NO_2^- and represent the mean ± SEM of *n* experiments run in triplicates.

■ ASSOCIATED CONTENT

Supporting Information

Figure S1, NMR data of *SSR*-**2**, Cartesian coordinates of **1** and **2**, copy of 1D and 2D NMR spectra of **1** and **2**, and two animations (avi) that show how the methylene and aromatic ^{1}H NMR signals of **1** change as temperature decreases. This material is available free of charge via the Internet at http://pubs.acs.org.

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