New Antimicrobial Flavanones from *Physena madagascariensis*

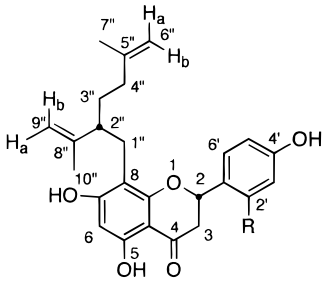
Yonghong Deng,[†] Jonathan P. Lee,[†] Manoelson Tianasoa-Ramamonjy,[†,‡] John K. Snyder,*[,†] Shelley Ann Des Etages,[§] Daniel Kanada,[§] Michael P. Snyder,[§] and Christopher J. Turner[⊥]

Department of Chemistry, Boston University, 590 Commonwealth Avenue, Boston, Massachusetts 02215, Department of Inorganic and Physical Chemistry, Faculty of Sciences, University of Antananarivo, P.O. Box 906, 101 Antananarivo, Madagascar, Department of Molecular, Cellular, and Developmental Biology, Yale University, New Haven, Connecticut 06520-8103, and Francis Bitter Magnet Laboratory, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139

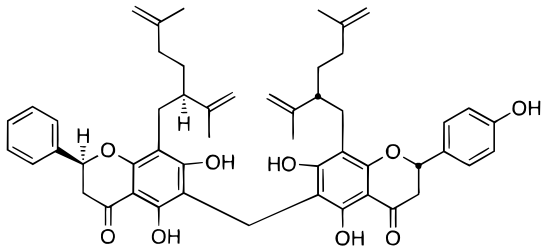
Received February 4, 2000

Two new flavanones (**1** and **2**) with antibacterial activity were isolated from the methanolic extract of the dried leaves of *Physena madagascariensis* using activity against *Staphylococcus aureus* to guide the isolation. A third flavonoid, a flavanone dimer linked by a methylene group (**3**) was also isolated and proved to be inactive. The structures of **1** and **2** were established primarily from NMR studies, while that of **3** required more extensive mass spectrometric analysis. All three flavanones had lavandulyl units in the limonene form. Flavanones **1** and **2** were active against several bacteria at concentrations as low as 4 μM.

Physena madagascariensis Noronha ex Thouars (Capparaceae),[1] known locally as "Remangily", is a tree endemic to Madagascar. The bark of the plant is rubbed on clothing as an effective repellent of terrestrial leeches, which are abundant in the region of the island where the tree is found.[2] We recently reported the isolation and structure of three new cytotoxic oleanane triterpene derivatives from the hexane-soluble fraction of the methanolic extract of the leaves of this plant. These triterpenes showed significant activity against human breast cancer cell lines.[3] Antibacterial and antifungal screens indicated that antimicrobial activity was prevalent in the 30% aqueous methanol-soluble fraction. This activity was traced to two flavanones, which we have called remangiflavanones A and B (**1** and **2**, respectively), whose structures are reported herein. Remangiflavanone C (**3**), a dimeric flavonoid possessing a methylene linkage between the two flavonoid monomeric subunits, was also isolated from both the aqueous methanol- and hexane-soluble fractions, but showed no activity in any bioassays (antimicrobial or cytotoxicity). All three compounds (**1**−**3**) represent previously unreported natural products with a lavandulyl monoterpene unit in the rare limonene form located at C-8.



1: R = H
2: R = OH



3

Results and Discussion

Repeated flash chromatography on Si gel of the 30% aqueous methanol-soluble fraction of the crude methanol extract[3] led to the isolation of **1** and **2** (see Experimental Section). The triphenolic flavanone structure of **1** (HREIMS *m*/*z* 408.1948, $[M]^+$, calcd for $C_{25}H_{28}O_5$, 408.1937) was readily apparent from the ^{1}H NMR (δ 11.99, 5.56, and 4.89, all exchangeable with D_2O, Table 1), ^{13}C NMR (δ 163.1, 161.8, and 155.9, Table 1: oxygenated aromatic carbons correlating with their respective hydroxyl protons in the HMBC spectrum, Supporting Information), and IR spectra (ν_{OH} 3450−3200 cm^{-1}). The low-field resonance of one of these phenolic groups (δ 11.99, OH-5), indicating intramolecular hydrogen bonding to a carbonyl group (^{13}C NMR, δ 196.8, C-4; IR, ν_{CO} 1636 cm^{-1}) along with an ABX pattern (δ 5.29, dd, J = 13.2, 2.9 Hz, H-2; 3.03, dd, J = 17.2, 2.9 Hz, H-3β; 2.77, dd, J = 17.2, 13.2 Hz, H-3α) are typical of 5-hydroxyflavanones.[4] Also immediately obvious in the ^{1}H NMR spectrum were two ortho-coupled aromatic doublets with a relative intensity of two protons each, suggestive of a para-substituted phenol as the B ring (δ 7.30 and 6.87, J = 8.8 Hz, H-2′/-6′ and H-3′/-5′, respectively), and a high-field aromatic singlet as commonly found in 1,3,5-trioxygenated, pentasubstituted A rings of flavanones (δ 5.98, H-6) with the corresponding carbons identified in the HMQC spectrum (δ 127.7, C-3′/-5′; 115.4, C-2′/-6′; 95.4, C-6). Benzylic/allylic cleavage[5] dominated the fragmentation of the molecular ion in the EIMS, giving the ions *m*/*z* 286 (61%) and 285 (100%, HREIMS *m*/*z* 285.0760, $[M - C_9H_{15}]^+$, calcd for $C_{16}H_{13}O_5$ 285.0763), with a subsequent retro-Diels−Alder fragmentation producing the ion at *m*/*z* 165 (23%) also supporting the assigned structure (Figure 1).[6]

The limonene form of the lavandulyl subunit with two sets of terminal olefinic protons (δ 4.56, 4.60, 4.62, 4.70,

* To whom correspondence should be addressed. Tel.: (617) 353-2621 (353-2575). Fax: (617) 353-6466. E-mail: jsnyder@chem.bu.edu.
[†] Boston University.
[‡] University of Antananarivo.
[§] Yale University.
[⊥] Massachusetts Institute of Technology.

10.1021/np000054m CCC: $19.00
Published on Web 06/30/2000

Table 1. ^{13}C and ^{1}H NMR Data for **1** and **2**

	1[a]		**2**[b]	
position	^{13}C (m)[c]	^{1}H (m, *J*)	^{13}C (m)[c]	^{1}H (m, *J*)
2	78.7 (d)	5.30 (dd, *J* = 13.2, 2.9 Hz)	75.4 (d)	5.67 (dd, *J* = 13.2, 2.9 Hz)
3α	43.1 (t)	3.03 (dd, *J* = 17.2, 13.2 Hz)	42.9 (t)	3.07 (dd, *J* = 17.2, 13.2 Hz)
3*β*		2.77 (dd, *J* = 17.2, 2.9 Hz)		2.76 (dd, *J* = 17.2, 2.9 Hz)
4	196.8 (s)		198.3 (s)	
4a	103.1 (s)		103.4 (s)	
5	161.8 (s)		163.1 (s)	
6	95.4 (d)	5.97 (s)	96.3 (d)	6.01 (s)
7	163.1 (s)		165.3 (s)	
8	107.0 (s)		107.8 (s)	
8a	160.6 (s)		162.2 (s)	
1′	130.8 (s)		117.9 (s)	
2′	127.7 (d)	7.31 (d, *J* = 8.8 Hz)	156.2 (s)	
3′	115.4 (d)	6.86 (d, *J* = 8.8 Hz)	103.5 (d)	6.48 (d, *J* = 2.2 Hz)
4′	155.9 (s)		159.5 (s)	
5″	115.4 (d)	6.86 (d, *J* = 8.8 Hz)	107.9 (d)	6.45 (dd, *J* = 8.4, 2.2 Hz)
6′	127.7 (d)	7.31 (d, *J* = 8.8 Hz)	128.8 (d)	7.39 (d, *J* = 8.4 Hz)
1″	27.7 (t)	2.56 (dd, *J* = 13.9, 7.0 Hz)	28.2 (t)	2.63 (d, *J* = 7.3 Hz, 2H)
1″		2.60 (dd, *J* = 13.9, 7.0 Hz)		
2″	46.5 (d)	2.30 (dddd, *J* = 7.0, 7.0, 7.3, 7.3 Hz)	47.2 (d)	2.52 (dddd, *J* = 7.3, 7.3, 7.3, 7.3 Hz)
3″	30.4 (t)	1.46 (ddd, *J* = 7.3, 7.7, 7.7 Hz, 2H)	30.6 (t)	1.49 (ddd, *J* = 7.3, 7.3, 7.3 Hz, 2H)
4″	35.5 (t)	1.89 (ddd, *J* = 14.6, 7.7, 7.7 Hz)	36.4 (t)	1.88 (ddd, *J* = 14.9, 7.3, 7.3 Hz)
4″		1.79 (ddd, *J* = 14.6, 7.7, 7.7 Hz)		1.82 (ddd, *J* = 14.9, 7.3, 7.3 Hz)
5″	146.2 (s)		146.9 (s)	
6a″	109.5 (t)	4.62 (br)	110.0 (t)	4.59 (br)
6b″		4.56 (br)		4.56 (br)
7″	22.5 (q)	1.59 (s)	22.6 (q)	1.58 (s)
8″	148.3 (s)		148.9 (s)	
9a″	111.5 (t)	4.70 (br)	111.7 (t)	4.64 (br)
9b″		4.60 (br)		4.60 (br)
10″	19.0 (q)	1.61 (s)	18.8 (q)	1.65 (s)
OH-5		11.99 (s)		12.19 (s)
OH-7		5.55 (s)		9.51 (s)
OH-2′				8.63 (s)
OH-4′		4.89 (s)		8.36 (s)

[a] Recorded in $CDCl_3$. [b] Recorded in acetone-d_6. [c] ^{13}C Multiplicities were determined by a DEPT experiment.

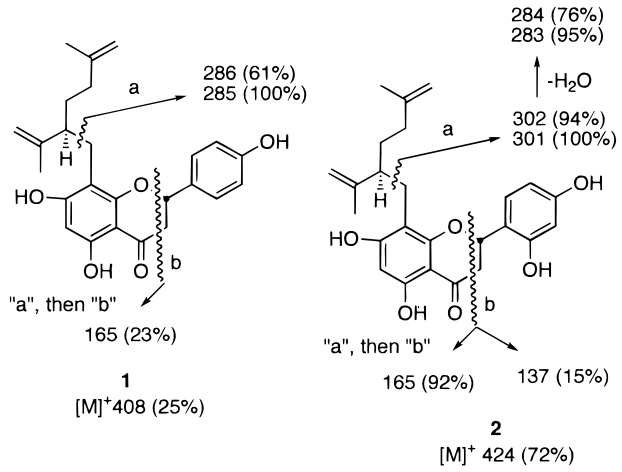
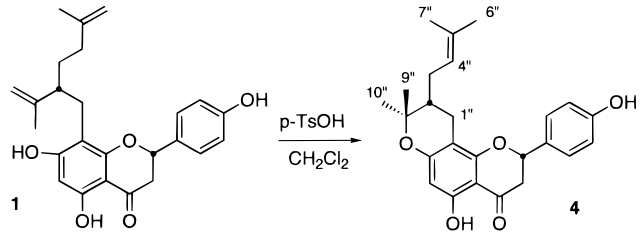


Figure 1. EIMS fragmentations of **1** and **2**. HREIMS confirmed fragment formulas for **2**.

all br s, 1H) was straightforward from the DQ−COSY (which revealed the $CH_2-CH-CH_2-CH_2$ spin system for H-1″ through H-4″), HMQC, and HMBC spectra (Supporting Information). The HMBC correlations (H-7″/C-4″, H-4″/C-7″, H-10″/C-2″, and H-2″/C-10″) allowed assignment of the two vinyl methyl and terminal methylene resonances, with difference NOE (DNOE) experiments then distinguishing the syn and anti vinyl protons at both C-6″ and C-9″ upon saturation of the vinyl methyls. Confirmation of the A ring substitution pattern was accomplished from the long-range heteronuclear couplings observed in the HMBC spectrum. The chelated phenolic proton showed couplings with the high-field aromatic methine carbon (C-6) and a high-field aromatic quaternary carbon (*δ* 103.1, C-4a), indicating that C-6 is unsubstituted and the site of the lavandulyl group is C-8.[7] A second phenolic proton (*δ* 5.56, OH-7) also showed coupling with C-6, as well as with another high-field aromatic quaternary carbon at *δ* 107.0 (C-8). The isolated aromatic singlet (H-6) also coupled with C-4a and C-8 as well as with C-5 and C-7, thereby confirming that the monoterpene substituent is located at C-8. Other long-range heteronuclear couplings (H-1″/C-8, C-7, C-8a and H-2″/C-8) were in accord with the proposed structure.

Scheme 1



The absolute stereochemistry at C-2 was routinely assigned as 2*S* from n→*π** (312 nm, Δ*ϵ* +1.00) and *π*→*π** (291 nm, Δ*ϵ* −5.81) bands in the CD spectrum,[8] considered the "natural" flavanone stereochemistry.[9] The stereochemistry at the C-2″ stereogenic center was more difficult to ascertain, and indeed is typically not assigned in lavandulylated flavanones. Acid-catalyzed closure (*p*-TsOH) of the pyran ring produced **4** (72%) with concurrent rearrangement of the terminal 5″,6″-olefin to the more stable trisubstituted double bond (Scheme 1). In this more rigid structure, the C-1″ methylene protons appeared as the AB

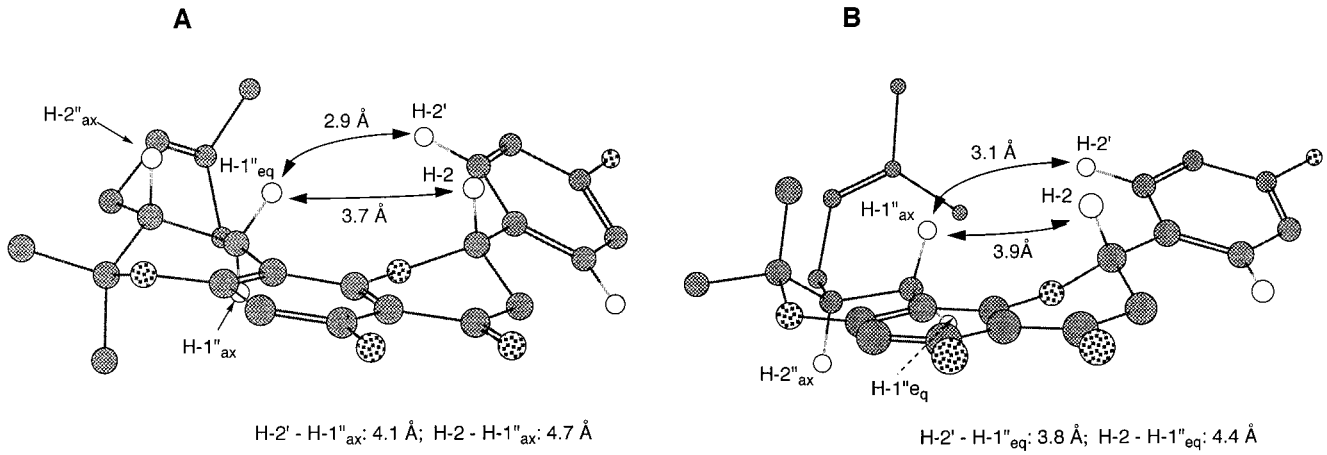


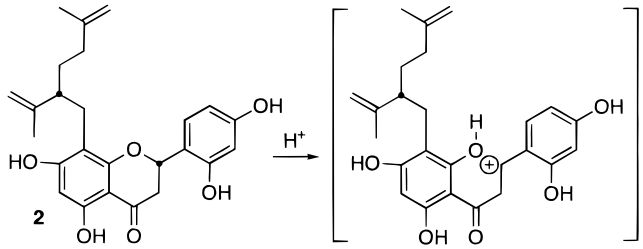
Figure 2. Conformations for (**A**) cyclic compound **4** with the (*R*)-C-2″ stereogenic center, and (**B**) the diastereomer with (*S*)-C-2″ center predicted by MM2 calculations and determined by NMR. Calculated distances from the H-2 and H-2′ protons to H-1″$_{eq}$ and H-1″$_{ax}$ for both diastereomers are given.

part of an ABX system (δ 2.69, J_{AB} = 16.9 Hz, J_{AX} = 5.3 Hz; 2.10 J_{AB} = 16.9 Hz, J_{AX} = 10.2 Hz), whose coupling constants with H-2″ indicated that the remaining prenyl side chain was equatorial (H-2″ axial). Distinction of the equatorial and axial H-1″ protons was also possible from these coupling constants with H-2″ (H-1″$_{ax}$ δ 2.10, H-1″$_{eq}$ δ 2.69). A DNOE experiment in $CDCl_3$ with irradiation of H-2 (δ 5.34, dd, J = 13.2, 2.9 Hz) showed a weak (0.3−0.5%) but reproducible enhancement of the equatorial H-1″, which therefore must be β-oriented. The same NOE was confirmed by a gradient-enhanced nuclear Overhauser effect spectroscopy (GOESY) spectrum[10] performed in benzene-d_6, where greater spectral dispersion of the H-3 and H-1″ protons was observed. This NOE indicates that C-2″ is an *R* stereogenic center; if this center had *S* stereochemistry, the predicted distance between H-2 and the H-1″ equatorial proton (α-oriented for an *S* center) would be 4.4 Å (MM2 calculations), an unlikely distance for the observation of an NOE with other relaxation pathways readily available. In contrast, the distance between H-2 and the H-1″ equatorial proton in **4** was calculated to be 3.7 Å, considerably more reasonable for the weak NOE observed (Figure 2).

Furthermore, in the GOESY spectrum, an NOE was also observed between the H-2′ protons of the B ring and H-1″$_{eq}$, though not with H-1″$_{ax}$. This observed NOE also supports the *R* stereochemistry for C-2″ inasmuch as the nearest approach of the H-2′ protons to H-1″$_{eq}$ is 2.9 Å, but 4.1 Å to H-1″$_{ax}$. With an *S* stereocenter at C-2″, the predicted distances from molecular modeling from the H-2′ protons would be 3.8 Å and 3.1 Å to H-1″$_{eq}$ and H-1″$_{ax}$, respectively. These latter distances are not compatible with the NOEs observed to H-2′.

With the structure of **1** assigned, **2** was readily identified as the 2′-hydroxyl derivative of **1** from the proton spectra (1D and DQ−COSY), which showed three aryl protons with a 1,2,4-trisubstituted aryl ring pattern of ortho and meta couplings (δ 6.45, dd, J = 8.3, 2.2 Hz, H-5′; 6.48, d, J = 2.2 Hz, H-3′; 7.39, d, J = 8.3 Hz, H-6′, Table 1) and carbon spectra (1D, DEPT, HMQC, and HMBC), which revealed two oxygenated carbons in the B ring (δ 156.2, C-2′; 159.5, C-4′,Table 1). In contrast to **1**, however, these NMR spectra had to be recorded in acetone-d_6 due to very limited solubility in $CDCl_3$, a consequence of the additional OH. The additional oxygen was also apparent from the HREIMS (*m/z* 424.1851, $[M]^+$, calcd for $C_{25}H_{28}O_6$, 424.1886) and LREIMS, which showed the benzylic/allylic fragmentation as in **1** (*m/z* 301, 100%, HREIMS *m/z* 301.0707, calcd for

Scheme 2



$C_{16}H_{13}O_6$, 301.0712) as well as the retro-Diels−Alder fragments 165 (91%, HREIMS *m/z* 165.0192, calcd for $C_8H_5O_4$, 165.0188) and 137 (15%, HREIMS *m/z* 137.0593, calcd for $C_8H_9O_2$, 137.0602, Figure 1). The fourth phenolic proton, easily identified by D_2O exchange (δ 8.63, OH-2′, Table 1), coupled with C-1′, C-2′, and C-3′ (HMBC), with the latter carbon bearing the meta-coupled proton doublet at δ 6.48, d, J = 2.2 Hz, H-3′. The downfield aryl proton showing only ortho-coupling (δ 7.39, d, J = 8.4 Hz, H-6′) also coupled with C-2 (δ 75.4) in the HMBC spectrum. These observations eliminated the possibility of a C-2′/C-5′ dioxygenated B-ring. The proton and carbon resonances of the A and C rings, as well as the monoterpene subunit, were nearly identical to those observed for **1** with the exception of the phenolic protons, whose shifts were sensitive to both concentration and the water content of the $CDCl_3$, and C-2, which was shifted upfield in **2** by 3.3 ppm as a result of the γ-effect due to the C-2′ hydroxyl group.

As with **1**, the configuration of the C-2 stereocenter in **2** was assigned as *S* from the CD spectrum (n→π*, 314 nm, $\Delta\epsilon$ +1.45; $\pi\rightarrow\pi$* 291 nm, $\Delta\epsilon$ −13.17), but attempts to apply the acid-catalyzed cyclization/NOE analysis strategy that was successful with **1** to assign the stereochemistry of the lavandulyl side chain of **2** met with failure. Treatment of **2** with acid led to racemization of the C-2 stereocenter prior to the desired cyclization. Presumably the greater stability of the intermediate benzylic carbocation due to the additional hydroxyl group on the aromatic B ring greatly accelerates C-ring opening with subsequent ring closure occurring to either face of the carbocation, resulting in the observed racemization of the C-2 stereocenter (Scheme 2).[11] On the basis of biogenetic considerations, we assume that the stereocenter of the lavandulyl side chain in **2** is also *R*, as determined for **1**, but this was not rigorously established.

In addition to **1** and **2**, a third, less polar flavanone was also isolated from both the 30% aqueous methanol-soluble

Table 2. ^{13}C and ^{1}H Data for **3**[a]

	monomer I		monomer II	
position	^{13}C (m)[b]	^{1}H (m, *J*)	^{13}C (m)[b]	^{1}H (m, *J*)
2	78.7 (d)	5.24 (dd, *J* = 12.6, 2.5 Hz)	78.9 (d)	5.32 (dd, *J* = 13.1, 2.5 Hz)
3	43.0 (t)[c]	3.04 (dd, *J* = 17.6, 12.6 Hz)	43.1 (t)[c]	3.04 (dd, *J* = 17.9, 13.1 Hz)
3		2.78 (dd, *J* = 2.5, 17.6 Hz)		2.83 (dd, *J* = 2.5, 17.9 Hz)
4	197.0 (s)		196.8 (s)	
4a	101.94 (s)[c]		101.91 (s)[c]	
5	157.04 (s)[c]		157.02 (s)[c]	
6	106.1 (s)[c]		106.0 (s)[c]	
7	163.2 (s)[c]		163.1 (s)[c]	
8	109.44[c]		109.38 (s)[c]	
8a	159.2 (s)[c]		159.1 (s)[c]	
1′	130.6 (s)		138.5 (s)	
2′	127.7 (d)	7.30 (d, *J* = 8.1 Hz)	125.9 (d)	7.40 (m)
3′	115.5 (d)	6.86 (d, *J* = 8.1 Hz)	128.7 (d)	7.40 (m)
4′	156.1 (s)		128.7 (d)	7.40 (m)
5′	115.5 (d)	6.86 (d, *J* = 8.1 Hz)	128.7 (d)	7.40 (m)
6′	127.7 (d)	7.30 (d, *J* = 8.1 Hz)	125.9 (d)	7.40 (m)
1″	27.8 (t)	2.62 (d, *J* = 8.4 Hz, 2H)	27.8 (t)	2.62 (2H, d, *J* = 8.4 Hz)
2″	46.36 (d)[c]	2.38 (dddd, *J* = 7.7, 7.7, 7.7, 7.7 Hz)	46.33 (d)[c]	2.38 (dddd, *J* = 7.7, 7.7, 7.7, 7.7 Hz)
3″	30.03 (t)[c]	1.42 (ddd, *J* = 7.7, 7.7, 7.7 Hz, 2H)	29.96 (t)[c]	1.42 (ddd, *J* = 7.7, 7.7, 7.7 Hz)
4″	35.46 (t)[c]	1.86 (ddd, *J* = 15.6, 7.7, 7.7 Hz)	35.42 (t)[c]	1.86 (ddd, *J* = 15.6, 7.7, 7.7 Hz)
4″		1.78 (ddd, *J* = 15.6, 7.7, 7.7 Hz)		1.78 (ddd, *J* = 15.6, 7.7, 7.7 Hz)
5″	146.40 (s)[c]		146.39 (s)[c]	
6a″	109.1 (t)	4.57 (br)	109.1 (t)	4.57 (br)
6b″		4.52 (br)		4.52 (br)
7″	22.57 (q)[c]	1.59 (s)[c]	22.55 (q)[c]	1.57 (s)[c]
8″	147.8 (s)		147.8 (s)	
9a″	111.2 (t)	4.63 (br)	111.2 (t)	4.63 (br)
9b″		4.53 (br)		4.53 (br)
10″	18.44 (q)[c]	1.62 (s)[c]	18.41 (q)[c]	1.61 (s)[c]
OH−5		13.64 (s)[c]		13.62 (s)[c]
OH−7		9.11 (s)[c]		9.09 (s)[c]
OH-4′		4.97 (s)		
CH_2	15.1 (t)	3.78 (br)		

[a] Recorded in $CDCl_3$. [b] ^{13}C multiplicities were determined by a DEPT experiment. [c] The assignments may be reversed between the two monomeric subunits.

and *n*-hexane-soluble fractions. The proton and carbon spectra indicated the dimeric nature of **3** from the "doubling" of most resonances (Table 2). Several key features of these spectra indicated that **3** was a nonsymmetric flavanone dimer linked through a methylene carbon. First, two prominent signals in the ^{1}H NMR spectrum were not "doubled": a methylene singlet (δ 3.78, s, 2H; ^{13}C δ 15.1) and a phenolic OH resonance (δ 4.97, s). In the proton spectrum, nine aromatic protons were indicated by integration, with only five distinct aromatic CH carbons appearing in the ^{13}C and HMQC spectra, indicating two distinct aryl B rings, each with symmetry. After assignment of the carbon resonances from the HMQC and HMBC experiments, it was apparent that one of these rings had typical *p*-hydroxyphenyl chemical shifts for the proton (δ 7.30, d, *J* = 8.1 Hz, H-2′/-6′; 6.86, d, *J* = 8.1 Hz, H-3′/-5′, Table 2) and carbon resonances, while the other B ring was otherwise unsubstituted (δ 7.35−7.45, m, 5H).

Conspicuously absent in the ^{1}H NMR spectrum were resonances typical for H-6 of a 5,7-dihydroxyflavanone, with the carbon resonances now indicating both flavanone subunits to be 6,8-disubstituted 5,7-dihydroxyflavanones. In the HMBC spectrum, coupling between the methylene protons (H-9) and C-5, C-6, and C-7 of both monomeric subunits was observed, indicating that the 6-position of each flavanone subunit was the site of dimerization through the methylene linker. Coupling between the chelated OH protons with the respective 6-position carbons (δ 13.54 with 106.1, and δ 13.62 with 106.0) confirmed that the two lavandulyl side chains, easily distinguished in the $^{1}H-^{1}H$ COSY spectrum as described above for **1** and **2**, were located at C-8 and C-8′ and not at C-6 and C-6′.

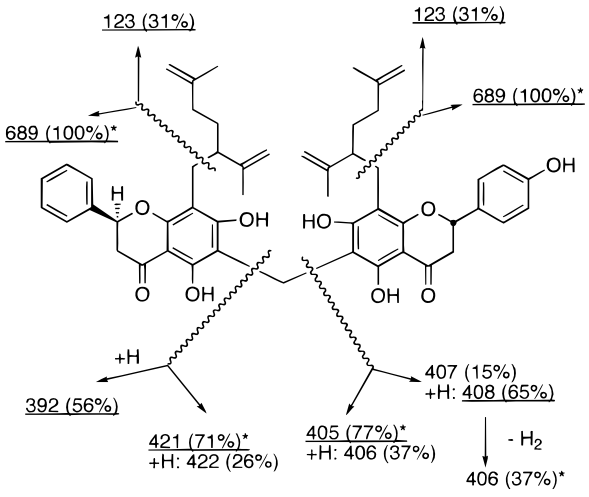


Figure 3. Main fragmentations of molecular ion of **3** in EIMS. Those pathways confirmed by detection of metastable ions by linked scanning with constant B/E are indicated by an asterisk (*). Fragments whose formulas were confirmed by HREIMS are underlined.

The mass spectrum further supported the assigned structure of **3** (HREIMS, *m/z* 812.3938, $[M]^+$, calcd for $C_{51}H_{56}O_9$, 812.3924). Loss of C_9H_{15} from either of the lavandulyl side chains from the molecular ion produces either of two fragment ions at *m/z* 689 (HREIMS, *m/z* 689.2734, $[M - C_9H_{15}]^+$, calcd for $C_{42}H_{41}O_9$, 689.2751, Figure 3), which is the base peak (EIMS, 100%). In addition, the molecular ion also showed cleavage about either side of the methylene linker producing the ions as shown, with the fragmentation to the benzylic ions (HREIMS, *m/z* 421.2052, calcd for $C_{26}H_{29}O_5$, 421.2015; 405.2072,

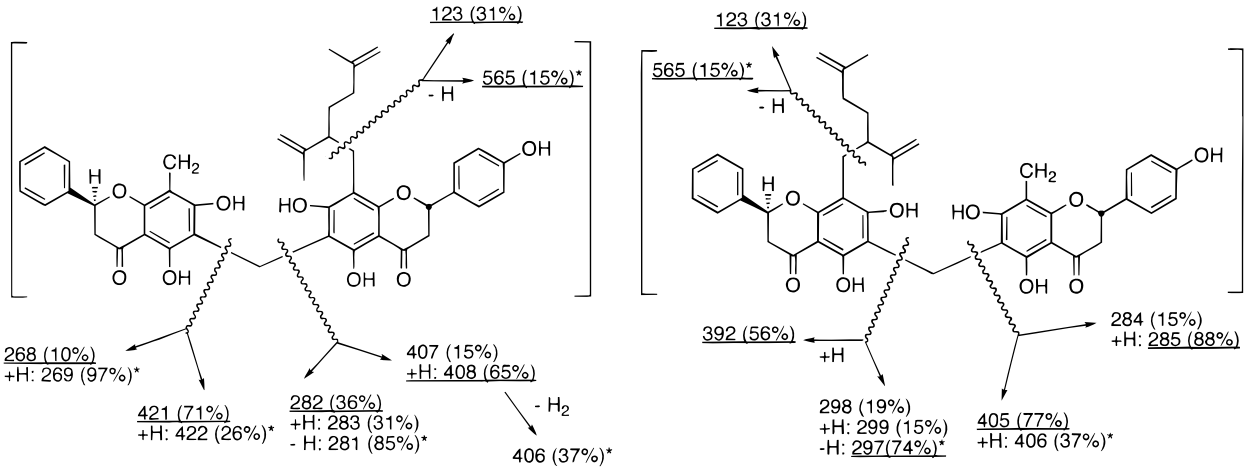


Figure 4. Fragmentations of the two ions composing the base peak in the EIMS of **3** at *m*/*z* 689. Those pathways confirmed by metastable ion detection by linked scanning with constant B/E are indicated by an asterisk (∗). Fragments whose formulas were confirmed by HREIMS are underlined.

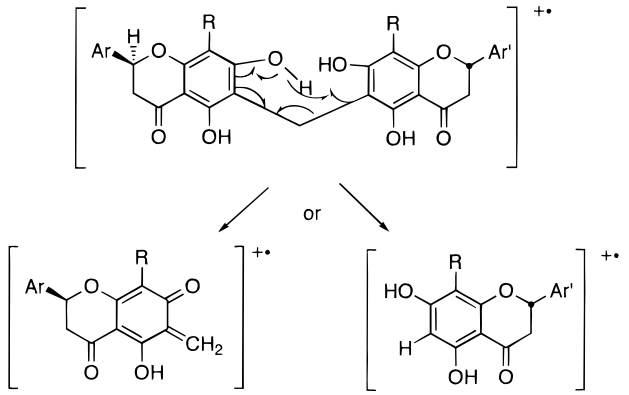


Figure 5. McLafferty rearrangement pathway for cleavage about the methylene linker in the EIMS of **3**.

calcd for $C_{26}H_{29}O_4$, 405.2066) confirmed by metastable ion detection through linked scanning with constant B/E.[12] Subsequent loss of the C_9H_{15} subunit from the remaining lavandulyl chain from either ion at *m*/*z* 689 produces the ion at 565 (HRMS, *m*/*z* 565.1526, calcd for $C_{33}H_{25}O_9$, 565.1499), with cleavage about the methylene linker from either ion at *m*/*z* 689 giving rise to several other predominant fragments observed in the mass spectrum (*m*/*z* 299, 298, 297, 285, 284, 283, 282, 281, and 268), all of which were supported by HREIMS data (see Experimental Section) and many also by linked scanning detection of metastable ions (Figure 4). One possible pathway for these cleavages about the methylene linker would be a McLafferty rearrangement (Figure 5), directly accounting for ions *m*/*z* 408/281, 392/297, 269, and 285, which was supported by HREIMS data.

Other typical flavanone fragmentations were also observed in accord with those discussed above for **1**. Thus, retro-Diels−Alder cleavages from all of the monomeric fragment ions were observed, confirmed by metastable ion detection by the linked scan at constant B/E (Figure 6). As with **1** and **2**, the absolute stereochemistry at C-2 and C-2′ was assigned as (2*S*, 2′*S*) from the CD spectrum (n→π*, 330 nm, $\Delta\epsilon$ +1.87; $\pi\rightarrow\pi$* 290 nm, $\Delta\epsilon$ −9.58), and the configurations of the lavandulyl side chains were assumed to have the *R* stereochemistry based on the assignment for **1**.

Flavanones **1** and **2** are closely related to other flavanones prenylated with lavandulyl subunits, such as the exiguaflavanones, the kushenols and kurarinols, and the sophoraflavanones all originally isolated from *Sophora* species,[5,7,13,14] although most previous reports of the occurrence of this monoterpene subunit in flavonoids indicate the terpinol isomeric form, not the limonene form as found here (Figure 7). Indeed, sophoraflavanone G (**5**)[7,13d] is identical with **2**, with the exception of the isomeric form of the lavandulyl subunit.[15] Lavandulol was originally isolated from French lavender oil by Schinz and Seidel as a mixture of two isomers, with the limonene isomeric form **6a** readily isomerizing into the terpinol isomeric form **6b**.[16] Subsequent synthetic studies from the same group confirmed the structures of the two lavandulol isomers,[17] and the absolute stereochemistry of the terpinol isomer was later shown to be the *R*-configuration by degradation studies,[18] and more recently confirmed by enantioselective synthesis.[19] Similar to the observation of Schinz and Seidel, acid-catalyzed closure of **1** to **4** in this work also resulted in the double-bond isomerization as found in the more common terpinol-form of lavandulol. Since then, a few other examples of the limonene form of the lavandulyl subunit in prenylated aromatic compounds from the Guttiferae family have been reported.[20] The *R*-stereocenter of **4** (and hence of **1**) corresponds to an *S*-stereocenter in lavandulol, which is antipodal to that found in lavender oil. A glycosidated (*S*)-lavandulol derivative has been reported, however, from *Hovenia dulcis*,[21] and both the (*R*)- and (*S*)-lavandulyl chains on Guttiferae aromatics have been confirmed by X-ray analysis.[20a,22]

In addition to the rarity of occurrence of the limonene-form of the lavandulyl monoterpene subunit, methylene-linked flavonoid dimers are also unusual. In the only other example found, Waterman and co-workers have reported a methylene-linked biflavanone from *Bosistoa brassii* with the linkage occurring between the C-6 and C-6′ positions of the monomeric flavanone subunits.[23] Examples of methylene-linked bicoumarins are also known in the literature.[24]

Flavanones **1**−**3** were tested against several Gram-positive and Gram-negative bacteria (Table 3). Both **1** and **2** were bacteriocidal against *S. aureus*, *S. epidermidis*, and *Enterococcus* sp. Remangiflavanone B (**2**) was also bacteriocidal against *Listeria monocytogenes*, while **1** was found to be bacteriostatic against this microorganism. In general, **2** was slightly more potent than **1**, with minimum effective concentrations (MECs) as low as 4 μM against *S. aureus*, *S. epidermidis*, and *Enterococcus* sp. Remangiflavanone C (**3**) was inactive against all organisms tested. The level of

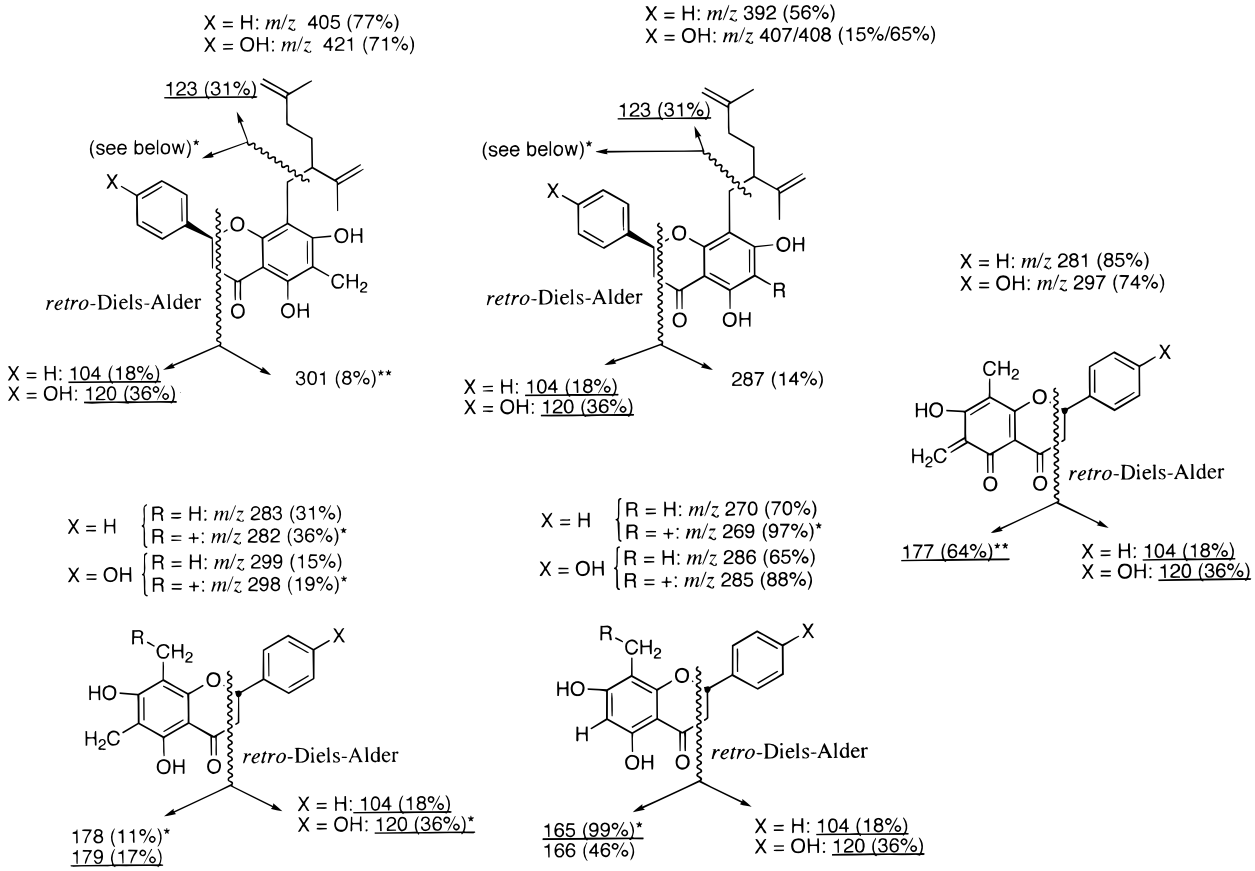


Figure 6. Fragmentations of monomeric subunit ions, dominated by benzylic cleavages and retro-Diels−Alder fragments. Those pathways confirmed by metastable ions detected by linked scanning with constant B/E are indicated by asterisks (∗). A double asterisk (∗∗) indicates metastable ions were detected from both R = H and R = OH subunits. Fragments whose formulas were confirmed by HREIMS are underlined.

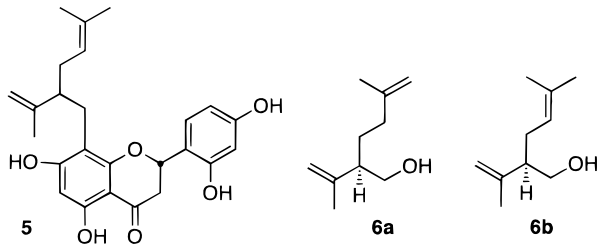


Figure 7. Structure of sophoraflavanone G (**5**)[7,13d] with the lavandulyl chain in the terpinol form, an olefinic isomer of flavanone **2**; and structures of the isomeric lavandulols reported by Schinz and Seidel,[16] **6a** with the limonene, and **6b** with the terpinol form.

activity of **1** and **2** is comparable to that observed with other flavanones against various microorganisms,[5,25] including that of exiguaflavones A and B.[25b]

Experimental Section

General Experimental Procedures. The ^{1}H NMR and ^{13}C NMR spectra of **1**, **3**, and **4** were recorded at 93.94 kG (^{1}H 400 MHz), 70.5 kG (^{13}C 75 MHz), and 63.41 kG (^{13}C 67.5 MHz) in $CDCl_3$ unless otherwise noted, at ambient temperature. The NMR spectra of **2** were recorded at the same field strengths in acetone-d_6. Chemical shifts are expressed in parts per million (ppm) relative to internal reference: for $CDCl_3$, the residual $CHCl_3$ resonance at δ 7.24 (^{1}H NMR) and the center line of the $^{13}CDCl_3$ triplet at δ 77.0 (^{13}C NMR); for $CD_3C(O)CD_3$, the residual $CHD_2C(O)CD_3$ resonance at δ 2.05 (^{1}H NMR) and the center line of the $^{13}CD_3C(O)CD_3$ septuplet at δ 29.9 (^{13}C NMR); for C_6D_6, the residual C_6HD_5 at δ 7.16 resonance (^{1}H NMR). All OH protons were confirmed by D_2O exchange. The HMQC spectra were optimized for an average $^1J_{C,H}$ of 135 Hz; the HMBC spectra were optimized for $J_{C,H}$ of 4.5 and 7 Hz. DNOE spectra of **4** were acquired with presaturation times of 8 s in repetitive blocks of 16 transients each for on-resonance (saturation) and off-resonance (negative) spectra using cyclic irradiation[26] to minimize the H_2 power. The GOESY spectra of **4** were run in benzene-d_6 in a 3-mm probe adapting the literature procedure[10b] using a mixing time of 1.25 s with a delay between sequences of 2.5 s. Mass spectra (LRMS and exact mass) were recorded on a Finnigan MAT-90 spectrometer as indicated. Metastable ion fragmentations were recorded in the EI mode by linked scanning with fixed B/E. IR spectra were recorded after depositing a $CDCl_3$ solution of the sample on an NaCl plate and evaporating the solvent. Melting points were determined on a capillary melting point apparatus and are uncorrected. All organic solvents were dried and distilled according to standard procedures[27] immediately before use. Flash chromatography was performed using flash Si gel (43−60 μm).[28]

Plant Material. The leaves of *P. madagascariensis* were collected and processed as previously described.[3]

Extraction and Isolation. Flavanones **1** and **2** were found in the 30% aqueous methanol fraction, which showed antimicrobial activity; dimer **3** was found in both the hexane-soluble fraction as well as the 30% aqueous methanol fraction.[3] The hexane-soluble fraction was not antimicrobial in these assays. The 30% aqueous methanol-soluble portion (9.5 g) of the methanol extract of the leaves was subjected to flash chromatography on Si gel eluting with a step gradient of acetone in hexanes producing nine fractions, F1−F9 (2% acetone, 200 mL, F1; 5% acetone, 600 mL, F2 and F3; 10% acetone, 550 mL, F4 and F5; 15% acetone, 200 mL, F6 and F7; 25% acetone, 250 mL, F7 and F8; and 50% acetone, 100 mL, F9). The third fraction (F3, 245 mg, eluted with 5% acetone) contained **3**, the fifth fraction (F5, 655 mg, eluted with 10% acetone) contained **1**, and the sixth fraction (F6, 510 mg, eluted with 15% acetone) contained **2**. Further flash chromatography on Si gel again

Table 3. Antibacterial Activity of **1** and **2**

species	**1** MEC, μg/mL, effect	**2** MEC, μg/mL, effect
Staphylococcus aureus	3.75 (9 μM), bacteriocidal	1.8 (4 μM), bacteriocidal
Staphylococcus epidermidis	1.8 (4 μM), bacteriocidal	1.8 (4 μM), bacteriocidal
Salmonella enteritidis	125 (0.3 mM), slow growth	125 (0.3 mM), bacteriostatic
Shigella sonnei	125 (0.3 mM), slow growth	125 (0.3 mM), slow growth
Escherichia coli	125 (0.3 mM), slow growth	125 (0.3 mM), slow growth
Enterobacter aerogenes	125 (0.3 mM), slow growth	125 (0.3 mM), slow growth
Listeris monocytogenes	125 (0.3 mM), slow growth	1.8 (4 μM), bacteriocidal
Enterococcus sp.	7.5 (18 μM), bacteriocidal	7.5 (18 μM), bacteriocidal

using step gradients of acetone in hexanes (5% acetone, 10% acetone, 20% acetone, and 40% acetone) of each of these fractions gave the pure compounds: **3** (87 mg; R_f 0.45, hexanes−acetone, 7:3) eluted with 10% acetone; **1** (294 mg; R_f 0.29, hexanes−acetone, 7:3) eluted with 20% acetone, and **2** (30 mg; R_f 0.26, hexanes−acetone, 7:3) eluted with 20% acetone.

Remangiflavanone A (1): yellow crystals; mp 147−149 °C; $[\alpha]^{24}_D$ −83.3° (*c* 0.18, MeOH); CD (*c* 1.08 mM, MeOH) 216 ($\Delta\epsilon$ +4.95), 235 (+1.61), 252 (+0.65), 291 (−5.81), 312 (+1.00) nm; UV (MeOH) λ_{max} (ϵ) 228 (18 010), 294 (11 760), 337 (3710) nm; IR (NaCl) ν_{max} 3348, 3080, 2965, 2934, 1636, 1600, 1518, 1438, 1374, 1345, 1248, 1174, 1072, 886, 832 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 1; EIMS (70 eV) *m*/*z* 408 (M^+, 25), 286 (61), 285 (100), 165 (23); HREIMS (70 eV) *m*/*z* 408.1948 (M^+, calcd for $C_{25}H_{28}O_5$, 408.1937), 285.0760 ([M − C_9H_{15}]$^+$, calcd for $C_{16}H_{13}O_5$, 285.0763).

Remangiflavanone B (2): yellow crystals; mp 168−170 °C; $[\alpha]^{24}_D$ −46.4° (*c* 0.14, MeOH); CD (*c* 1.06 mM, MeOH) 226 ($\Delta\epsilon$ +9.19), 255 (+0.83), 291 (−13.17), 314 (+1.45) nm; UV (MeOH) λ_{max} (ϵ) 228 (20 470), 292 (19 640), 337 (4090) nm; IR (NaCl) ν_{max} 3376, 3080, 2975, 2940, 1635, 1604, 1525, 1444, 1385, 1355, 1300, 1275, 1255, 1175 1158, 1100, 1072, 890, 835 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 1; EIMS (70 eV) *m*/*z* 425 ([M + 1]$^+$, 23), 424 (M^+, 72), 302 (94), 301 (100), 284 (76), 283 (95), 166 (21), 165 (91), 137 (15), 69 (29); HREIMS (70 eV) *m*/*z* 424.1851 (M^+, calcd for $C_{25}H_{28}O_6$, 424.1886), 301.0707 ([M − C_9H_{15}]$^+$, calcd for $C_{16}H_{13}O_6$, 301.0712), 283.0617 ([M − C_9H_{15} − H_2O]$^+$, calcd for $C_{16}H_{11}O_5$, 283.0606), 166.0270 (calcd for $C_8H_6O_4$, 166.0266), 165.0192 (calcd for $C_8H_5O_5$, 165.0188), 137.0593 (calcd for $C_8H_9O_2$, 137.0603).

Remangiflavanone C (3): yellow crystals; mp 104−106 °C; $[\alpha]^{24}_D$ −56.8° (*c* 0.19, MeOH); CD (*c* 0.66 mM, MeOH) (+0.74), 290 (−9.58), 330 (+1.87) nm; UV (MeOH) λ_{max} (ϵ) 232 (35 980), 300 (25 250), 337 (14 120) nm; IR (NaCl) ν_{max} 3339, 3080, 2970, 2950, 1633, 1535, 1480, 1443, 1375, 1345, 1280, 1198, 1175, 1124, 887, 840 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 2; EIMS (70 eV) *m*/*z* 812 (M^+, 8), for relevant fragment ions, see Figures 3, 4, and 6; HREIMS (70 eV) *m*/*z* 812.3938 (M^+, calcd for $C_{51}H_{56}O_9$, 812.3924), 689.2734 ([M − C_9H_{15}]$^+$, calcd for $C_{42}H_{41}O_9$, 689.2751), 565.1526 (calcd for $C_{33}H_{25}O_9$, 565.1499), 421.2052 (calcd for $C_{29}H_{29}O_5$, 421.2015), 408.1934 (calcd for $C_{25}H_{28}O_5$, 408.1937), 405.2072 (calcd for $C_{26}H_{29}O_4$, 405.2066), 392.1985 (calcd for $C_{25}H_{28}O_4$, 392.1988), 297.0791 (calcd for $C_{17}H_{13}O_5$, 297.0763), 285.0746 (calcd for $C_{16}H_{13}O_5$, 285.0763), 282.0868 (calcd for $C_{17}H_{14}O_4$, 282.0892), 268.0749 (calcd for $C_{16}H_{12}O_4$, 268.0736), 179.0332 (calcd for $C_9H_7O_4$, 179.0344), 177.0171 (calcd for $C_9H_5O_4$, 177.0188), 165.0202 (calcd for $C_8H_5O_4$, 165.0188), 123.1174 (calcd for C_9H_{15}, 123.1174), 120.0578 (calcd for C_8H_8O, 120.0575), 104.0607 (calcd for C_8H_8, 104.0626).

Acid-Catalyzed Cyclization of 1. To a solution of **1** (9 mg, 0.022 mmol) in anhydrous CH_2Cl_2 (2.5 mL) was added *p*-TsOH (1.9 mg, 0.011 mmol). The mixture was stirred at room temperature for 16 h, then the reaction was quenched by the addition of saturated $NaHCO_3$ solution (2 mL). The aqueous and organic layers were separated, and the aqueous layer extracted with CH_2Cl_2 (3 × 2 mL). The combined organic layers were dried over Na_2SO_4, filtered, and the solvent removed in vacuo. The crude residue was purified by flash chromatography (hexanes−acetone, 85:15) to yield **4** (5 mg, 0.012 mmol, 55%, 72% based on recovered **1**) as a white solid: mp 84−86 °C; $[\alpha]^{24}_D$ −148.0° (*c* 0.1, MeOH); IR (NaCl) ν_{max} 3394, 2977, 2930, 1641, 1592, 1519, 1486, 1450, 1340, 1313, 1270, 1169, 1128, 1092, 833 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.75 (1H, s, OH-5), 7.32 (2H, d, *J* = 8.4 Hz, H-2′/H-6′), 6.88 (2H, d, *J* = 8.4 Hz, H-3′/H-5′), 5.95 (1H, s, H-6), 5.34 (1H, dd, *J* = 13.2, 2.9 Hz, H-2), 5.13 (1H, dd, *J* = 7.1, 7.1 Hz, H-4″), 4.90 (1H, s, OH-4′), 3.00 (1H, dd, *J* = 17.1, 13.2 Hz, H-3α), 2.77 (1H, dd, *J* = 17.1, 2.9 Hz, H-3β), 2.70 (1H, dd, *J* = 16.9, 5.3 Hz, H-1″β), 2.22 (1H, m, H-3″), 2.11 (1H, dd, *J* = 16.9, 10.2 Hz, H-1″α), 1.76 (1H, m, H-3″), 1.68 (3H, s, H-6″), 1.64 (1H, m, H-2″), 1.56 (3H, s, H-7″), 1.40 (3H, s, H-9″), 1.18 (3H, s, H-10″); ^{1}H NMR (400 MHz, C_6D_6) δ 12.66 (1H, s, OH-5), 6.92 (2H, d, *J* = 8.2 Hz, H-2′/H-6′), 6.51 (1H, s, H-6), 6.42 (2H, d, *J* = 8.2 Hz, H-3′/H-5′), 5.00 (1H, dd, *J* = 6.8, 6.8 Hz, H-4″), 4.72 (1H, dd, *J* = 12.8, 2.9 Hz, H-2), 3.96 (1H, s, OH-4′), 2.78 (1H, dd, *J* = 16.8, 5.1 Hz, H-1″β), 2.54 (1H, dd, *J* = 16.8, 12.8 Hz, H-3α), 2.33 (1H, dd, *J* = 16.8, 2.9 Hz, H-3β), 2.18 (1H, dd, *J* = 16.8, 10.3 Hz, H-1″α), 2.02 (1H, m, H-3″), 1.60 (2H, m, H-2″/H-3″), 1.53 (3H, s, H-6″), 1.40 (3H, s, H-7″), 1.21 (3H, s, H-9″), 1.01 (3H, s, H-10″); ^{13}C NMR (100 MHz, $CDCl_3$) δ 195.8 (C-4), 162.7 (C-7), 161.4 (C-5), 159.7 (C-8a), 155.8 (C-4′), 133.2 (C-5″), 131.2 (C-1′), 127.6 (C-2′/C-6′), 122.1 (C-4″), 115.5 (C-3′/C-5′), 102.6 (C-4a), 101.4 (C-8), 97.2 (C-6), 79.6 (C-8″), 78.5 (C-2), 43.2 (C-3), 40.9 (C-2″), 29.4 (C-3″), 27.6 (C-9″), 25.8 (C-6″), 21.9 (C-1″), 20.9 (C-10″), 17.9 (C-7″); EIMS (70 eV) *m*/*z* 409 ([M + 1]$^+$, 13), 408 (M^+, 5), 394 (18), 393 (81), 285 (20), 273 (14), 219 (15), 165 (40), 120 (26), 69 (15), 28 (100); HREIMS (70 eV) *m*/*z* 408.1913 (M^+, calcd for $C_{25}H_{28}O_5$, 408.1937); unreacted **1** was also recovered (2 mg, 22%).

Bioassays. YPAD [yeast extract (1%), peptone (2%), dextrose (2%), and adenine (40 μg/mL) with 95% water] was used as a permissive medium.[29] YPAD was inoculated with bacteria (see Table 3) using a sterile pipet tip and incubated overnight (16 h) at 24 °C. The wells of a microtiter plate were filled with 200 μL of YPAD containing 0, 125.0, 62.0, 31.0, 7.5, 3.75, or 1.8 μg/mL of the test compound, and inoculated with 5 μL of the overnight culture. The microtiter plates were then incubated overnight (16 h) and scored visually. In the cases where no growth was observed, cells from the well were spread on YPAD plates (YPAD and 2% agar) and incubated overnight (16 h) again at room temperature. The absence of growth indicated that the compound was bacteriocidal at the test concentration, while growth indicated that the compound was bacteriostatic at the test concentration. Slow growth in the microtiter wells was noted as such.

Acknowledgment. We are grateful to the Boston University Community Technology Fund for financial support. We also thank Dr. Etienne A. Rakotobe, Department of Ethnobotany, National Center for Pharmaceutical Research, Antananarivo, Madagascar, for identification of *P. madagascariensis*.

Supporting Information Available: Structurally definitive spectra for **1**−**3**, tables of HMBC and NOE correlations for **1**−**3**, and ^{1}H and ^{13}C NMR spectra for **4**. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) *Physena* spp. have been recently suggested to be more appropriately entered into a distinct Physenaceae family: Morton, C. W.; Karol, K. G.; Chase, M. W. *Bot. Rev.* **1997**, *63*, 231−239.

(2) Sibree, J. *A Naturalist in Madagascar*; Seeley, Service, & Co.: London, 1915.
(3) Deng, Y.; Jiang, T.-Y.; Sheng, S.; Tianasoa-Ramamonjy, M.; Snyder, J. K. *J. Nat. Prod.* **1999**, *62*, 471−476.
(4) For a review of the proton spectra of flavonoids including flavanones: Markham, K. R.; Mabry, T. J. In *The Flavonoids*; Harborne, J. B., Mabry, T. J., Mabry, H., Eds.; Academic: New York, 1975; Part I, pp 62−75.
(5) Kuroyanagi, M.; Arakawa, T.; Hirayama, Y.; Hayashi, T. *J. Nat. Prod.* **1999**, *62*, 1595−1599.
(6) For a review of the mass spectra of flavonoids including flavanones: Mabry, T. J.; Markham, K. R. In *The Flavonoids*; Harborne, J. B., Mabry, T. J., Mabry, H., Eds.; Academic: New York, 1975; Part I, pp 78−126.
(7) Historically, the location of such prenyl side chains has caused difficulties that have been resolved by the observation of long-range heteronuclear couplings to the chelated OH: Shirataki, Y.; Yokoe, I.; Noguchi, M.; Tomimori, T.; Komatsu, M. *Chem. Pharm. Bull.* **1988**, *36*, 2220−2225.
(8) (a) Gaffield, W. *Tetrahedron* **1970**, *26*, 4093−4108. (b) Bohm, B. A. In *The Flavonoids*; Harborne, J. B., Mabry, T. J., Mabry, H., Eds.; Academic: New York, 1975; Part I, pp 594−595.
(9) (a) Bohm, B. A. In *The Flavonoids: Advances in Research*; Harborne, J. B., Mabry, T. J., Eds.; Chapman and Hall: New York, 1982; pp 349−350. (b) Bohm, B. A. In *The Flavonoids: Advances in Research Since 1980*; Harborne, J. B., Ed.; Chapman and Hall: New York, 1988; p 348.
(10) (a) Stonehouse, J.; Adell, P.; Keeler, J.; Shaka, A. J. *J. Am. Chem. Soc.* **1994**, *116*, 6037−6038. (b) Stott, K.; Stonehouse, J.; Keeler, J.; Hwang, T.-L.; Shaka, A. J. *J. Am. Chem. Soc.* **1995**, *117*, 4199−4200.
(11) Shimokoriyama, M. *J. Am. Chem. Soc.* **1957**, *79*, 4199−4202.
(12) (a) Boyd, R. K.; Beynon, J. H. *Org. Mass Spectrom.* **1977**, *12*, 163−164. (b) Millington, D. S.; Smith, J. A. *Org. Mass Spectrom.* **1977**, *12*, 264−265. (c) Bruins, A. P.; Jennings, K. R.; Evans, S. *Int. J. Mass Spectrom. Ion Phys.* **1978**, *26*, 395 404. For a review: (d) Jennings, K. R.; Mason, R. S. In *Tandem Mass Spectrometry*; McLafferty, F. W., Ed.; Wiley-Interscience: New York, 1983; pp 197−222.
(13) (a) Hatayama, K.; Komatsu, M. *Chem. Pharm. Bull.* **1971**, *19*, 2126−2131. (b) Kyogoku, K.; Hatayama, K.; Komatsu, M. *Chem. Pharm. Bull.* **1973**, *21*, 2733−2738. (c) Wu, L. J.; Miyase, T.; Ueno, A.; Kuroyanagi, M.; Noro, T.; Fukushima, S. *Chem. Pharm. Bull.* **1985**, *33*, 3231−3236. (d) Iinuma, M.; Tanaka, T.; Mizuno, M.; Shirataki, Y.; Yokoe, I.; Komatsu, M.; Lang, F. A. *Phytochemistry* **1990**, *29*, 2667−2669. (e) Ruangrungsi, N.; Iinuma, M.; Tanaka, T.; Ohyama, M.; Yokoyama, J.; Mizuno, M. *Phytochemistry* **1992**, *31*, 999−1001. (f) Iinuma, M.; Yokoyama, J.; Ohyama, M.; Tanaka, T.; Mizuno, M.; Ruangrungsi, N. *Phytochemistry* **1993**, *33*, 203−208. (g) Iinuma, M.; Yokoyama, J.; Ohyama, M.; Tanaka, T.; Ruangrungsi, N. *Phytochemistry* **1994**, *35*, 785−789.
(14) For a review of prenylated flavonoids: (a) Barron, D.; Ibrahim, R. K. *Phytochemistry* **1996**, *43*, 921−982. For older reviews of flavonoids including flavanones, see refs 8b and 9.
(15) The structure of sophoraflavanone G (**5**) was originally assigned to norkurarinone (ref 13a), but this latter flavanone was later assigned as the C-6 prenylated flavanone (ref 7). Vexibinol, which was originally assigned as this C-6 prenylated flavanone, was then also assigned as **5**.[7]
(16) Schinz, H.; Seidel, C. F. *Helv. Chim. Acta* **1942**, *25*, 1572−1591.
(17) Grütter, H.; Schinz, H. *Helv. Chim. Acta* **1952**, *35*, 1656−1660.
(18) Soucek, M.; Dolejs, L. *Collect. Czech. Chem. Commun.* **1959**, *24*, 3802−3803.
(19) Kramer, A.; Pfander, H. *Helv. Chim. Acta* **1982**, *65*, 293−301.
(20) (a) Blount, J. F.; Williams, T. H. *Tetrahedron Lett.* **1976**, 2921−2924. (b) delle Monache, F.; delle Monache, G.; Bettolo, G. B. M.; Lyra, D. A.; Lwande, W. *Gazz. Chim. Ital.* **1984**, *114*, 55−60. (c) Gustafson, K. R.; Blunt, J. W.; Munro, M. H. G.; Fuller, R. W.; McKee, T. C.; Cardellina, J. H.; McMahon, J. B.; Cragg, G. M.; Boyd, M. R. *Tetrahedron* **1992**, *48*, 10093−10102. (d) Seo, E.-K.; Wall, M. E.; Wani, M. C.; Navarro, H.; Mukherjee, R.; Farnsworth, N. R.; Kinghorn, A. D. *Phytochemistry* **1999**, *52*, 669−674.
(21) Yoshikawa, K.; Nagai, M.; Wakabayashi, M.; Arihara, S. *Phytochemistry* **1993**, *34*, 1431−1433. In this paper the authors drew the (*R*)-lavandulyl subunit in the structure but state in the text that the correct stereochemistry should be (*S*) and give optical rotation for the isolated lavandulol as the *S*-(+)-enantiomer. Presumably the drawing of the (*R*)-lavandulol was an oversight.
(22) Krishnamurthy, N.; Ravindranath, B.; Guru Row, T. N.; Venkatesan, K. *Tetrahedron Lett.* **1982**, 2233−2236.
(23) Parsons, I. C.; Gray, A. I.; Waterman, P. G.; Hartley, T. G. *J. Nat. Prod.* **1993**, *56*, 46−53.
(24) Basa, S. C. *Phytochemistry* **1988**, *27*, 1933−1941.
(25) (a) Iinuma, M.; Tsuchiya, H.; Sato, M.; Yokoyama, J.; Ohyama, M.; Ohkawa, Y.; Tanaka, T.; Fujiwara, S.; Fujii, T. *J. Pharm. Pharmacol.* **1994**, *46*, 892−895. (b) Chanphen, R.; Thebtaranonth, Y.; Wanauppathamkul, S.; Yuthavong, Y. *J. Nat. Prod.* **1998**, *61*, 1146−1147.
(26) Kinns, M.; Sanders, J. K. M. *J. Magn. Reson.* **1984**, *56*, 518−520.
(27) Perrin, D. D.; Armarego, W. L. F.; Perrin, D. R. *Purification of Laboratory Chemicals*, 2nd ed.; Pergamon: New York, 1980.
(28) Still, W. C.; Kahn, M.; Mitra, A. *J. Org. Chem.* **1978**, *43*, 2923−2925.
(29) Sherman, F. In *Methods in Enzymology*; Guthrie, C., Fink, G. R., Eds.; Academic: New York, 1991; Vol. 194, Chapter 1.

NP000054M