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AN ANTIFUNGAL BIBENZYL FROM THE NEW ZEALAND LIVERWORT, *PLAGIOCHILA STEPHENSONIANA*. BIOACTIVITY-DIRECTED ISOLATION, SYNTHESIS, AND ANALYSIS

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ABSTRACT.—The bioactivity-directed isolation of 4-hydroxy-3′-methoxybibenzyl [**1**] from a New Zealand liverwort, *Plagiochila stephensoniana*, is described. Compound **1** exhibited antifungal and antibacterial activity. Compound **1** is not considered to be a useful taxonomic marker. Syntheses of **1**, stilbenes **2** and **3**, and some derivatives were made and the biological activities were compared.

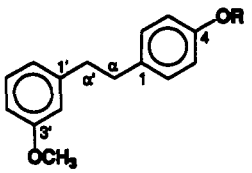
There is considerable interest in bryophytes, especially liverworts, as sources of biologically active compounds (1). However, the biological activity of the rich bryophyte flora in New Zealand's temperate rain forests has not been explored. Therefore, we have included samples of mosses and liverworts in our program of screening New Zealand plants and fungi for new, biologically active, natural products.

One of the liverwort extracts that showed antifungal activity was from *Plagiochila stephensoniana* Mitt. (Plagiochilaceae, order Jungermanniales). This is the most robust of the 26 *Plagiochila* species recognized in New Zealand; it forms fronds up to 25 cm tall (2). It is generally found at low altitudes in the South Island but at altitudes up to 1000 m in the North Island. It is also found in Tasmania (2). There are no reports of medicinal uses for *Pl. stephensoniana* (3); indeed, very few liverworts have had medicinal or physiological activities reported (1). Previous studies on a North Island collection found that the major Et_2O-soluble component was 4-hydroxy-3′-methoxybibenzyl [**1**], along with low levels of some mono- and sesquiterpenes (α- and β-pinene, alloaromadendrene, bicyclogermacrene), 3,4′-dimethoxybibenzyl, and two unidentified bibenzyls (4). A new verrucosane diterpene and a sesquiterpene, spathulenol, have been isolated as minor components (5). No biological activities were reported for any of these compounds.

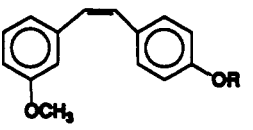
We now report the bioactivity-directed isolation of **1** as the major antifungal component of *Pl. stephensoniana*. Bibenzyl **1** and related compounds have been synthesized to study the structural basis of this antifungal activity. Three other New Zealand *Plagiochila* species contained **1** at much lower levels.

Screening assays showed that a small-scale extract of *Pl. stephensoniana* was active against the dermatophyte fungus *Trichophyton mentagrophytes*, the yeast *Candida albicans*, and the Gram-positive bacteria *Bacillus subtilis* (Table 1). Preliminary chromatographic studies on this extract showed good recovery of the antifungal activity in medium and low polarity fractions from octadecyl-bonded reversed-phase (C_{18} Rp) and Si gel columns. Activity against *T. mentagrophytes* was used to direct the isolation of the major active component. A larger-scale extract was chromatographed over a C_{18} (reversed-phase) column, with the most active fraction further chromatographed over Sephadex LH 20 and finally over Si gel to give a single active compound. This was identified as the known compound, 4-hydroxy-3′-methoxybibenzyl [**1**], by comparison with published ^{1}H-nmr, uv and ir spectra (4,6).

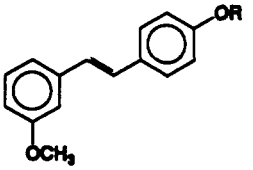
Disk assays on **1** (at 60 μg/disk) showed activity against *B. subtilis* and *Ca. albicans* as well as *T. mentagrophytes*, but not against the Gram-negative bacteria *Escherichia coli* and



1 R=H
6 R=SiMe$_2$*t*-Bu
7 R=Me



2 R=H
4 R=SiMe$_2$*t*-Bu



3 R=H
5 R=SiMe$_2$*t*-Bu

Pseudomonas aeruginosa (Table 2). Bibenzyl **1** also showed cytotoxicity against monkey kidney cells (BSC) at 60 μg/well, but it did not inhibit the cytopathic effects of either Herpes simplex Type I or Polio Type I viruses (7). Compound **1** did not inhibit the growth of P-388 leukemia cells (IC_{50} >25.0 μg/ml).

There is only one literature report associating bibenzyl **1** with bioactivity (6). In this report, **1** showed activity against the brown rot fungi, *Gloeophyllum trabeum* and *Poria placenta* (IC_{50} 87 and 11 μg/ml). In the same study other bibenzyls were found to be active against the brown rot fungi and also the white rot fungus, *Coriolus versicolor.* Stilbenes, closely related to bibenzyls, are important factors in the durability of wood (8).

This present report, therefore, represents the discovery of new biological activities associated with bibenzyl **1** and *Pl. stephensoniana* from which it was isolated. The bibenzyl was found to exhibit activity down to 12 μg/disk in the disk bioassays (Table 2). Dilution assays gave minimum inhibitory concentration and minimum fungicidal activity results against *T. mentagrophytes* and *Ca. albicans* for **1** of 125 μg/ml and 62.5 μg/ml, respectively (Table 3).

Bibenzyl **1** has been reported in other liverworts (9) including one *Plagiochila* species (10), so it was possible that the antimicrobial activity of our other *Plagiochila* extracts (Table 1) could be due to this compound. Therefore, we checked by hplc for the presence of **1** in our four other *Plagiochila* collections.

The survey showed large variation in the levels of **1** within our *Plagiochila* collections

TABLE 1. Antimicrobial Activities and Bibenzyl Levels of *Plagiochila* Extracts.

Species	Herbarium No.	*Bacillus subtilis*[a]	*Candida albicans*[a]	*Trichophyton mentagrophytes*[a]	Bibenzyl levels[b]
Plagiochila fasciculata	OTA 046556	1	nd[c]	7	trace[d]
Plagiochila stephensoniana	OTA 046557	4	6	12	10.5
Plagiochila banksiana	OTA 046558	5	5	1	0.05
Plagiochila suborbiculata	OTA 046567	2	1	nd	nd
Plagiochila deltoidea	OTA 046568	nd	1	nd	0.54

[a]Activity is represented as the width of the zone of inhibition in millimeters.
[b]Mg of bibenzyl **1** per g dry liverwort.
[c]None detected.
[d]Unable to resolve peak.

TABLE 2. Biological Activities of Prepared Compounds in Disk Diffusion Assays.

Sample	Dose μg/disk	Cytotoxicity[a]	Antibacterial[b]		Antifungal[b]		
			Escherichia coli	*Bacillus subtilis*	*Candida albicans*	*Trichophyton mentagrophytes*	*Cladosporium resinae*
1	2.4			inactive	inactive	inactive	
	12			3	2	7	
	60	ww	inactive	5	10	7	2
	300			5	10	7	
2	2.4			inactive	inactive	5	
	12			4	2	7	
	60	+	1	9	7	10	3
	300			9	7	10	
3	2.4			inactive	inactive	2	
	12			2	inactive	8	
	60	+	inactive	4	2	6	2
	300			6	7	10	
4	60	inactive	inactive	inactive	inactive	inactive	inactive
	300			inactive	inactive	inactive	inactive
5	60	inactive	inactive	inactive	inactive	inactive	inactive
	300			inactive	inactive	inactive	inactive
6	60	inactive	inactive	inactive	inactive	inactive	inactive
	300			inactive	inactive	inactive	inactive
7	60	inactive	inactive	inactive	inactive	inactive	inactive
	300			inactive	inactive	inactive	
(*E*)-Stilbene	60		inactive	inactive	inactive	inactive	inactive
m-Cresol	60	inactive	inactive	inactive	inactive	2	inactive
	300			inactive	inactive	inactive	
Methyl anisole	60	inactive	inactive	inactive	inactive	inactive	inactive
	300			inactive	inactive	inactive	
Reference[c]			8	12	10	8	10

[a]Cytotoxicity was measured against monkey kidney cells (BSC) represented as zone sizes, +, ++, +++, and ww (whole well).

[b]Activity is represented as the width of the zone of inhibition in mm.

[c]Reference compounds and doses: antifungal, nystatin (100 units/disk); *E. coli*, gentamicin (10 μg/disk); *B. subtilis*, chloramphenicol (30 μg/disk).

(Table 1). The level in *Pl. stephensoniana* was very high and corresponded to approximately 1% of the dry wt of the plant or 30% of the EtOH extract. *Plagiochila deltoidea* had the second highest level of **1**, but only one-twentieth the level in *Pl. stephensoniana*.

TABLE 3. Biological Activities of Prepared Compounds in Dilution Assays.

Sample	*Candida albicans*		*Trichophyton mentagrophytes*		P388[a]
	MIC[b]	MFC[c]	MIC	MFC	IC_{50}
1	125.00	125.00	62.50	62.50	>25.00
2	31.25	62.50	15.62	15.62	16.30
3	31.25	125.00	31.25	31.25	9.90
6	>1000[d]	>1000	>1000	>1000	>25.00
Reference[e]	31.25	31.25	62.50	62.50	0.02

[a]Concentration in μg/ml to give 50% inhibition of P-388 cells.

[b]Minimum inhibitory concentration in μg/ml.

[c]Minimum fungicidal concentration in μg/ml.

[d]No inhibition detected at 1000 μg/ml.

[e]Reference compounds: MIC and MFC, nystatin; P-388, mitomycin C.

The extract of *Pl. deltoidea* had correspondingly low antimicrobial activity (Table 1). The other three species contained very low levels of bibenzyl **1**, so their antimicrobial activities must be due to other compounds. To our knowledge, none of these species have been investigated as yet.

Neither the occurrence nor the amount of **1** shows a close relation to the subgeneric classification of *Plagiochila* in New Zealand (11). It occurs in both the primary groups (the Cauliflorae and the Ramiflorae) of that classification, and the amount of bibenzyl varies within these groups. Bibenzyl **1** is also known from a number of other liverworts, including both leafy (Jungermannialean) (12) and thallose (Metzgerialean) (10) orders. The distribution of the bibenzyl, both within *Plagiochila* and among major groups of liverworts, suggests that it is of no taxonomic value.

Bibenzyl derivatives are the most characteristic compounds of liverworts and occur in all orders within the Hepaticae, but not in mosses or hornworts and only sporadically in higher plants (9). The most widely distributed bibenzyl derivative is lunularic acid (3,4′-dihydroxybibenzyl-2-carboxylic acid), the biogenetic precursor of **1**, which has been reported to cause inhibition of seed germination, root formation, and growth of roots, lettuce hypocotyls, and *Avena* coleoptiles (13). In a study of the inhibitory activity of bibenzyl derivatives in liverwort gemmaling and cress root growth assays, the two most active compounds were 3-hydroxy-4′-methoxybibenzyl and 4-hydroxybibenzyl, both closely related to **1** (which was not tested) (13). An allelopathic role has also been suggested for lunularic acid (9). It seems probable that the high level of **1** in *Pl. stephensoniana* (Table 1) will defend against fungal infections and may also have an allelopathic role. This may be related to the success of *Pl. stephensoniana*, the most robust of the New Zealand *Plagiochila* species (2).

Syntheses of bibenzyls have been accomplished by a number of methods (9). We chose the Wittig condensation of a benzyl phosphonium salt with a benzaldehyde. The use of NaH as the base in the condensation gave both the *Z* and *E* stilbenes in approximately equal amounts, in contrast to bases such as butyl lithium which give predominately one isomer (14). We desired both isomers for structural activity studies. Wittig condensation (15) of *m*-methoxybenzyltriphenylphosphonium bromide with *p*-*tert*-butyldimethylsilyloxybenzaldehyde gave a mixture of protected stilbenes. These were separated to give pure samples of the *Z* and *E* isomers **4** and **5**. Deprotection with tetra-*n*-butylammonium chloride and KF (16) afforded the *Z* and *E* stilbenes **2** and **3**. This is the first report of the *Z* isomer; data for the *E* isomer were consistent with literature values (17,18). Hydrogenation of a mixture of the stilbene isomers in the presence of Pd-C yielded the bibenzyl whose spectral data and chromatographic and bioactivity behavior were identical to those of the natural bibenzyl **1**. Protected bibenzyl **6** was prepared by treatment with *tert*-butyldimethylchlorosilane (19). Methylation of **1** gave **7** (4).

Stilbenes **2**–**5**, bibenzyls **1**, **6**, and **7**, (*E*)-stilbene, *m*-cresol, and methyl anisole were submitted to a range of in vitro biological assays (Tables 2 and 3). None of the compounds tested showed any activity against the Herpes simplex Type I or Polio Type I viruses or the bacteria *Pseudomonas aeruginosa*. Furthermore, compounds **4**–**7** along with *E*-stilbene, *m*-cresol, and methyl anisole exhibited no activity in the disk diffusion assays (Table 2) at levels up to 300 μg per disk.

These results, though limited, show some trends. A free OH group seems to be necessary for biological activity as has been found in other studies (6,8,20,21). However, *m*-cresol showed very little biological activity, despite its free OH group. This implies that the diaryl structure of the bibenzyl and stilbenes is important for biological activity. The dilution assays (Table 3) showed that bibenzyl **1** was less active than the *Z* and *E* stilbenes **2** and **3** against both *Ca. albicans* and *T. mentagrophytes*.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—All solvents were distilled before use and were removed by rotary evaporation at temperatures up to 45°. Reversed-phase flash chromatography followed the published method (22), and Davisil, 35–70 μm, 150 Å was used for Si gel flash chromatography. Tlc was carried out using Merck DC-Plastikfolien Kieselgel 60 F_{254}, visualized with a uv lamp. Mass, uv, and ir (film) spectra were recorded on Varian Mat CH-7 or Kratos MS80, Shimadzu UV 240, and Perkin-Elmer 1600 spectrometers, respectively. Nmr spectra of $CDCl_3$ solutions at 25° were recorded at 200 MHz for ^1H and 50 MHz for ^{13}C on a Varian Gemini-200 spectrometer. Chemical shifts are given in ppm on the δ scale referenced to the solvent peaks $CHCl_3$ at 7.25 and $CDCl_3$ at 77.00. For antimicrobial disk assays the sample solutions were dried onto 6-mm filter paper disks which were then placed onto seeded agar Petri dishes and incubated. Activity showed as a zone of inhibition around the disk, with its width recorded in mm. Antifungal assays for minimum inhibitory concentration were carried out using a standard tube assay method (23). For the antitumor assay a 2-fold dilution series of the sample was incubated for 72 h with P-388 (murine leukemia) cells. The concentration of the sample required to reduce the P-388 cell growth to 50% of the growth of a solvent control was determined using the absorbance obtained upon staining with MTT tetrazolium. Results were expressed as an IC_{50} in μg/ml. For the antiviral assay, samples of interest were dried onto 6-mm filter paper disks and placed directly onto BSC-1 cells (African Green Monkey Kidney) and infected with either Herpes simplex Type I virus (ATCC VR 733) or Polio Type I virus (Pfizer vaccine strain). After incubation for 24 h, assays were examined using an inverted microscope. Activity showed as a zone of viral and or cytotoxic inhibition around the disk (7). [Test organisms *Ca. albicans* and *T. mentagraphytes* are deposited in the culture collection of Otago University Microbiology (collection strain nos. 10261 and 10100).]

COLLECTION AND EXTRACTION.—*Pl. stephensoniana, Plagiochila banksiana,* and *Plagiochila fasciculata* were collected from the Dunedin (Leith Valley) area on the South Island of New Zealand in December 1991 [University of Otago Herbarium (OTA) specimen numbers 046557, 046558 and 046556]. *Plagiochila suborbiculata* and *Pl. deltoidea* were collected from Fiordland (Borland road) on the South Island of New Zealand in May 1992 (OTA 046567 and 046568). Oven-dried (50°) material was frozen with liquid N_2 and ground. Initial screening (Table 1) was carried out using 30 μl/disk of an extract produced by shaking this material (5.0 g) overnight in EtOH (50 ml).

BIOACTIVITY-DIRECTED ISOLATION OF 4-HYDROXY-3′-METHOXYBIBENZYL [**1**].—*Pl. stephensoniana* (10 g, ground) was extracted with EtOH (3×50 ml) by homogenizing and filtering to give a green/brown gum [0.316 g, Tm 10 @ 200 (i.e., *T. mentagrophytes* inhibition zone 10 mm at 200 μg/disk)]. Reversed-phase flash chromatography over C_{18} (0.315 g, precoated on 0.300 g C_{18}, loaded on a 3.8 g C_{18} column) eluted in steps from H_2O to $CHCl_3$, led to elution of the most active fraction (0.067 g, Tm 15 @ 200)with MeOH-$CHCl_3$ (3:1). Chromatography of this fraction over Sephadex LH 20 (5 g column) developed with MeOH-$CHCl_3$ (1:1) gave an active fraction (0.035 g, Tm >7 @ 150), a subsample (0.023 g) of which was purified over Si gel ($CHCl_3$) to give **1** as a pale brown oil (0.019 g): ^{13}C nmr 159.55 (C-3′), 153.71 (C-4), 143.55 (C-1′), 133.99 (C-1), 129.58 (C-2, -6), 129.31 (C-5′), 121.01 (C-6′), 115.19 (C-3, -5), 114.29 (C-2′), 111.29 (C-4′), 55.21 (OMe), 38.23 (C-α′), 36.91 (C-α).

ANALYSES OF LIVERWORTS FOR 4-HYDROXY-3′-METHOXYBIBENZYL.—Each liverwort sample was dried and ground, then a subsample (0.500 g) was homogenized (Ultra Turrax) for 1 min in MeOH (5 ml) containing (*E*)-stilbene (100 μg/ml) as internal standard. After standing for 5 min, an aliquot (0.5 ml) was filtered through a short C_{18} reversed-phase column (0.25 g C_{18}). After washing with MeOH (0.5 ml), the eluent was made up to 1 ml with MeOH and filtered (0.45 μm). Subsamples (20 μl) were analyzed on a Phenomenex Bondclone 10 C18 column (300×3.9 mm) with MeOH-H_2O (80:20) as mobile phase at a flow rate of 1.0 ml/min. Detection at 280 nm showed the peak for **1** at 5.7 min and the peak for *E*-stilbene at 10.2 min. Assignment of peaks was confirmed by co-injection of authentic **1** with the extracts.

(*Z*)- AND (*E*)-4-(*TERT*-BUTYLDIMETHYLSILOXY)-3′-METHOXYSTILBENES.—3-Methoxybenzyltriphenylphosphonium bromide was prepared by methylation of *m*-cresol (1.0 ml) with dimethylsulfate (8.8 g) and anhydrous K_2CO_3 (9.7 g) by refluxing for 4 h in dry Me_2CO (50 ml; yield 90%), followed by bromination with *N*-bromosuccinimide (24). The resulting bromide was treated with triphenylphosphine in DMF (4). Spectral data were consistent with those found in literature (4).

NaH (0.1 g; 60% suspension in oil) in a flame-dried flask was washed with pentane (3×2 ml), evacuated to dryness, and the flask filled with N_2. After cooling to 0°, dry THF (2 ml) and 3-methoxybenzyltriphenylphosphonium bromide (0.896 g) were added and the mixture stirred for 3.5 h. 4-*tert*-Butyldimethylsiloxybenzaldehyde [0.482 g, prepared from *p*-hydroxybenzaldehyde (19)] in THF (3 ml) was then added, the ice bath removed, and the mixture stirred for 48 h. H_2O (30 ml) was added and the

reaction mixture extracted with Et_2O (3×30 ml). The combined Et_2O fractions were washed with H_2O and brine and dried over anhydrous Na_2SO_4. Evaporation gave a gum (0.503 g) with the *Z* and *E* isomers **4** and **5** in 1:1 ratio by ^1H nmr. Cc over Si gel (15% $CHCl_3$/hexane) afforded the 4-*tert*-butyldimethylsiloxy-3′-methoxystilbenes in three fractions: pure *Z*, mixed *Z* and *E*, and pure *E*, in a total yield of 42%.

Z *Isomer* **4**.—Ir 2940, 1600, 1500, 1250, 1170, 910, 840, 770 cm^{-1}; ^1H nmr 0.17 (6H, s, Si-Me), 0.96 (9H, s, SiC-Me), 3.60 (3H, s, OMe), 6.46 (1H, d, J=12 Hz, H-α′), 6.52 (1H, d, J=12 Hz, H-α), 6.69 (2H, d, J=8 Hz, H-3, -5), 7.12 (2H, d, J=8 Hz, H-2, -6), 6.65–7.19 (4H, m, H-2′, -4′, -5′, -6′); ^{13}C nmr 159.33 (C-3′), 154.82 (C-4), 138.85 (C-1′), 130.26 (C-1), 130.12 (C-2, -6), 130.07 (C-5′), 129.16 (C-α), 128.71 (C-α′), 121.44 (C-6′), 119.77 (C-3, -5), 113.56 (C-4′), 113.18 (C-2′), 54.95 (OMe), 25.65 (SiC-Me), 18.19 (Si-C), −4.45 (Si-Me); hreims *m/z* $[M]^+$ 340.1861 (100%) ($C_{21}H_{28}O_2Si$ requires 340.1858), 283.1173 (79%) ($C_{17}H_{19}O_2Si$ requires 283.1154).

E *Isomer* **5**.—Ir 2930, 1600, 1510, 1270, 1170, 910, 840, 780 cm^{-1}; ^1H nmr 0.21 (6H, s, Si-Me), 0.99 (9H, s, SiC-Me), 3.84 (3H, s, OMe), 6.83 (2H, d, J=9 Hz, H-3, -5), 6.94 (1H, d, J=16 Hz, H-α′), 7.04 (1H, d, J=16 Hz, H-α), 7.38 (2H, d, J=9 Hz, H-2, -6), 6.75–7.54 (4H, m, H-2′, -4′ -5′, -6′); ^{13}C nmr 159.81 (C-3′), 155.41 (C-4), 139.04 (C-1′), 130.54 (C-1), 129.47 (C-5′), 128.53 (C-α), 127.64 (C-2, -6), 126.59 (C-α′), 120.23 (C-3, -5), 118.92 (C-6′), 112.76 (C-4′), 111.52 (C-2′), 55.02 (OMe), 25.62 (SiC-Me), 18.15 (Si-C), −4.47 (Si-Me); hreims *m/z* $[M]^+$ 340.1859 (100%) ($C_{21}H_{28}O_2Si$ requires 340.1858), 283.1149 (65%) ($C_{17}H_{19}O_2Si$ requires 283.1154).

(Z)-*4-Hydroxy-3′-methoxystilbene* [**2**].—Stilbene **4** (0.176 g), tetrabutylammonium chloride (0.344 g), anhydrous KF (0.068 g), H_2O (35 μl), and MeCN (2 ml) were stirred overnight, H_2O (2 ml) was added, and the solution was extracted with $CHCl_3$ (3×2 ml). The combined extracts were washed with H_2O and brine, dried over Na_2SO_4, and evaporated. Silica cc (20% EtOAc/hexane) gave **2** (0.082 g, 70%) as an oil: ir 3390, 1590, 1510, 1260 cm^{-1}; uv λ max (EtOH) 307 (ε 23,500), 322 (24,400), (EtOH/NaOH) 322 (25,000), 350 (32,400); ^1H nmr 3.68 (3H, s, OMe), 6.47 (1H, d, J=12 Hz, H-α′), 6.51 (1H, d, J=12 Hz, H-α), 6.67 (2H, d, J=9 Hz, H-3, -5), 7.14 (2H, d, J=9 Hz, H-2, -6), 6.56–7.22 (4H, m, H-2′, -4′, -5′, -6′); ^{13}C nmr 159.29 (C-3′), 154.64 (C-4), 138.87 (C-1′), 130.38 (C-2, -6), 129.92 (C-5′), 129.76 (C-1), 129.23 (C-α), 128.68 (C-α′), 121.47 (C-6′), 115.04 (C-3, -5), 113.78 (C-4′), 113.01 (C-2′), 54.95 (OMe); hreims *m/z* $[M]^+$ 226.0995 (100%) ($C_{15}H_{14}O_2$ requires 226.0994).

(E)-*4-Hydroxy-3′-methoxystilbene* [**3**].—Compound **3** was prepared via the above procedure from stilbene **5** as white crystals (71%). Spectral data were consistent with literature (17,18).

4-Hydroxy-3′-methoxybibenzyl [**1**].—Hydrogenation of a mixture of *Z* and *E* stilbenes (0.265 g) in EtOAc (5 ml) over Pd-C (5%) (0.200 g) gave, after filtration through celite and evaporation, **1** as an oil (0.260 g; 97%), identical spectroscopically to the natural product.

*4-(*tert*-Butyldimethylsiloxy)-3′-methoxybibenzyl* [**6**].—Imidazole (0.074 g) was added to a stirred solution of bibenzyl **1** (0.100 g) and *tert*-butyldimethylchlorosilane (0.080 g) in DMF (0.5 ml), and the mixture was stirred overnight. After dilution with 10% $NaHCO_3$ (10 ml) and extraction with hexane (3×10 ml), the combined extracts were washed with H_2O, dried over Na_2SO_4, and evaporated to give **6** (0.147 g, 98%) as an oil: ir 2930, 1600, 1510, 1490, 1260, 1170, 1150, 1050, 910, 840, 780 cm^{-1}; ^1H nmr 0.20 (6H, s, Si-Me), 1.00 (9H, s, SiC-Me), 2.87 (4H, s, H-α, -α′), 3.79 (3H, s, OMe), 6.63 (2H, d, J=9 Hz, H-3, -5), 7.17 (2H, d, J=9 Hz, H-2, -6), 6.69–6.81 (3H, m, H-2′, -4′, -6′), 7.14–7.25 (1H, m, H-5′); ^{13}C nmr 159.57 (C-3′), 153.73 (C-4), 143.52 (C-1′), 134.42 (C-1), 129.26 (C-2, -6), 129.20 (C-5′), 120.89 (C-6′), 119.85 (C-3, -5), 114.20 (C-2′), 111.20 (C-4′), 55.09 (OMe), 38.16 (C-α′), 37.01 (C-α), 25.71 (SiC-Me), 18.19 (Si-C), −4.43 (Si-Me); hreims *m/z* $[M]^+$ 342.2016 (25%) ($C_{21}H_{30}O_2Si$ requires 342.2015) $[M-C_8H_9O]^+$ 221.1354 (100%) ($C_{13}H_{21}OSi$ requires 221.1362), 121.0626 (14%) (C_8H_9O requires 121.0653).

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