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A NEW BIPHENYL DERIVATIVE FROM AN UNIDENTIFIED MARINE FUNGUS E33

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*A new biphenyl derivative 3,5′-dihydroxy-4′,5-dimethoxy-2′-methyl-[1,1′-biphenyl]-2-carboxylic acid methyl ester (**1**), was isolated from the culture broth of a fungus isolated from the rhizosphere soil of mangrove root on the South China Sea. The structure of **1** was determined by comprehensive spectroscopic methods, primarily 2D NMR techniques.*

Key words: mangrove, marine fungus, biphenyl.

Marine microorganisms have been recognized as a new source for the production of bioactive secondary metabolites. In recent years, numerous metabolites possessing uncommon structures and potent bioactivities have been isolated from strains of bacteria and fungi collected from diverse marine environments, such as animals, plants, and sediments. During the course of our search for biologically active substances from marine microorganisms from the South China Sea, we have obtained a number of interesting compounds [1-3]. In this paper, a new biphenyl derivative, 3,5′-dihydroxy-4′,5-dimethoxy-2′-methyl-[1,1′-biphenyl]-2-carboxylic acid methyl ester (**1**), was isolated from an unidentified fungus E33 collected from the rhizosphere soil of mangrove root on the South China Sea.

A 200 L fermentation broth was concentrated and extracted with ethyl acetate. The extract was repeatedly chromatographed on silica gel to obtain compound **1** from the 10% ethyl acetate/petroleum fraction as colorless crystals. Compound **1** has the molecular formula $C_{17}H_{18}O_6$ determined by HR-EIMS (*m/z* 318.1100 $[M]^+$, calcd for $C_{17}H_{18}O_6$, 318.1098) and NMR spectra. The IR spectrum showed absorbance for hydroxyl (3400 cm^{-1}) and ester carbonyl (1656 cm^{-1}) groups. Analysis of the ^{13}C NMR spectrum (Table 1) showed one carbonyl carbon signals (δ 171.3), twelve olefinic carbon signals (δ 145.7, 105.3, 264.4, 100.1, 163.7, 110.7, 135.4, 126.3, 111.6, 145.2, 142.7, 114.5), etc. These accounted for seven of the nine degrees of unsaturation required by the molecular formula, thereby showing that illustrating compound **1** is bicyclic. The ^1H NMR signals at δ 6.47 (1H, d, J = 2.4 Hz, H-6′), δ 6.22 (1H, d, J = 2.4 Hz) and δ 6.64 (1H, s), δ 6.67 (1H, s) showed that the rings should be two tetrasubstituted benzenes. Additionally, three methoxy groups at δ 3.48 (3H, s), 3.82 (3H, s), 3.91 (3H, s) and one methyl at δ (3H, s) along with two hydroxyls at δ 11.52 (s, 1H), 5.45 (s, 1H) were also found.

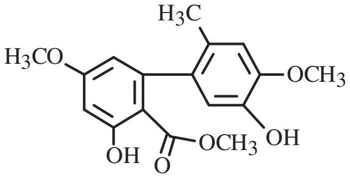

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TABLE 1. NMR Data for **1** ($CDCl_3$, TMS)

C atom	^1H NMR	^{13}C NMR (DEPT)	^1H-^1HCOSY	HMBC
1		145.7 (C)		
2		105.3 (C)		
3		164.4 (C)		
4	6.47 (d, 2.4 Hz, 1H)	100.1 (CH)	H-6	C-2, 3, 5, 6
5		163.7 (C)		
6	6.22 (d, 2.4 Hz, 1H)	110.7 (CH)	H-4	C-1′, 2, 4, 5
7		171.3 (C)		
8	3.48 (s, 3H)	51.7 (CH_3)		C-7
9	3.82 (s, 3H)	55.4 (CH_3)		C-5
1′		135.4 (C)		
2′		126.3 (C)		
3′	6.67 (s, 1H)	111.6 (CH)	H-7′	C-1′, 5′, 7′
4′		145.2 (C)		
5′		142.7 (C)		
6′	6.64 (s, 1H)	114.5 (CH)		C- 1′, 2′, 4′, 5′
7′	1.97 (s, 3H)	19.4 (CH_3)	H-3′	C-1′, 2′, 3′
8′	3.91 (s, 3H)	56.0 (CH_3)		C-4′
a-OH	11.52 (s, 1H)			C-2, 3, 4
b-OH	5.45 (s, 1H)			C-4′, 5′, 6′

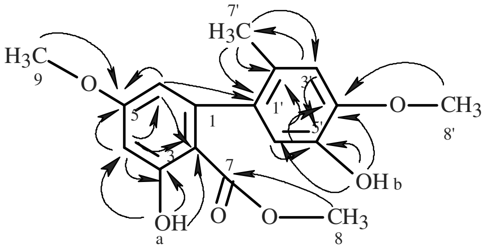

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Fig. 1. The selective correlations of the HMBC of **1**.

The assignment of protons attached to their corresponding carbons was readily accomplished by the HMQC technique. The correlations from H-8 and C-7 in HMBC showed the presence of an a-$COCH_3$ group. The positions of -$COCH_3$ (C-7, C-8), a-OH, and -OCH_3 (C-9) were determined by the values of chemical shifts and HMBC correlations from a-OH to C-2,3,4; H-4 to C-2,3,5,6; H-9 to C-5; H-6 to C-2,4,5 and ^1H-^1HCOSY correlations between H-4 to H-6. Similarly, the positions of -CH_3 (C-7′), b-OH, and -OCH_3(C-8′) were assigned by the values of chemical shifts and HMBC correlations from b-OH to C-4′,5′,6′; H-3′ to C-1′,5′,7′; H-8′ to C-4′; H-6′ to C-2′,4′,5′ and H-7′ to C-1′,2′,3′. Additionally the correlations from H-6 to C-1′; H-6′ to C-1′; H-7′ to C-1′ in HMBC confirmed the connectivity of C-1 and C-1′. Thus the overall structure of **1** was unambiguously established. Figure 1 shows the selective correlations of the HMBC of **1**.

EXPERIMENTAL

Melting points were detected on a Fisher-Johns hot-stage apparatus and are uncorrected. NMR data were recorded on a Varian Inova-500 NB spectrometer, using $CDCl_3$ as solvent and TMS as internal standard. Mass spectra were obtained on a VG-ZAB mass spectrometer, and HRMS on a VG Autospec-500 mass spectrometer. IR spectra were obtained on a Nicolet 5DX-FTIR spectrophotometer, and UV spectra were measured on a Shimadzu UV-240 spectrophotometer. Chromatography was carried out on silica gel column (200-300 mesh; Qingdao Haiyang chemicals).

Fungus Material and Culture Conditions. A strain of the fungus E33 was isolated from the South China Sea coast. It is apospory and its general species has not been identified. Starter cultures were maintained on cornmeal seawater agar. Plugs of agar supporting mycelial growth were cut and transferred aseptically to a 250 mL Erlenmeyer flask containing 100 mL of liquid medium (glucose 10g/L, Peptone 2 g/L, yeast extract 1 g/L, NaCl 30 g/L). The flask was incubated at 30°C on a rotary shaker for 5-7 days. The mycelium was aseptically transferred to 500 mL Erlenmeyer flasks containing culture liquid (200 mL). The flasks were then incubated at 30°C for 25 days.

Extraction and Separation of Metabolites. The cultures (200 L) were filtered through cheesecloth. The filtrate was concentrated to 5 L in vacuo below 50°C and extracted five times by shaking with an equal volume of ethyl acetate. The combined extracts were chromatographed repeatedly on a silica gel column using gradient elution from petroleum to ethyl acetate to obtain 3,5′-dihydroxy-4′,5-dimethoxy-2′-methyl-[1,1′-biphenyl]-2-carboxylic acid methyl ester (**1**) from the ethyl acetate-petroleum ether (15:85) fraction.

3,5′-Dihydroxy-4′,5-dimethoxy-2′-methyl-[1,1′-biphenyl]-2-carboxylic acid methyl ester (1): $C_{17}H_{18}O_6$, colorless needles, mp153-155°C, HREIMS 318.1100 (cacl. 318.1098), EIMS:318, 286, 271, 257, 243 , IR bands (neat, ν_{max}, cm^{-1}):3400, 2995, 2937, 2843, 1656, 1616, 1571, 1514, 1428, 1331, 1258, 1198, 1148, 953; ^{1}H NMR (500 MHz $CDCl_3$) and ^{13}C NMR data: see Table 1.

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REFERENCES

1. Y. C. Lin, X. Y. Wu, S. Feng, G. C. Jiang, S. N. Zhou, L. L. P. Vrijmoed, and E. B. Gareth Jones, *Tetrahedron Lett.,* **42**, 449 (2001).
2. Y. C. Lin, X. Y. Wu, S. Feng, G. C. Jiang, G. C. Luo, S. N. Zhou, L. L. P. Vrijmoed, E. B. Gareth Jones, K. Kroth, and K. Steingrover, F. Zsila, *J. Org. Chem.*, **66**, 6252 (2001).
3. G. Y. Chen, Y. C. Lin, L. Wen, L. L. P. Vrijmoed, and E. B. Gareth Jones, *Tetrahedron*, **59**, 4907 (2003).