Two New Mexicanolides from the Fruit of the Chinese Mangrove *Xylocarpus granatum*

Jun Wu[a], Si Zhang[a], Yang Song[b], Zhihui Xiao[a], Qiang Xiao[b], and Qingxin Li[a]

[a] Guangdong Key Laboratory of Marine Materia Medica, South China Sea Institute of Oceanology, Chinese Academy of Sciences, 164 West Xingang Road, Guangzhou 510301, P. R. China
[b] Institute of Organic Chemistry, Jiangxi Science & Technology Normal University, Nanchang 330013, P. R. China

Reprint requests to Dr. Jun Wu. Fax: +86-20-84451672. E-mail: wwujun2003@yahoo.com

Z. Naturforsch. **60b,** 1291 – 1294 (2005); received August 5, 2005

Two new mexicanolides, 3-deacetyl xyloccensin M and 3-deacetyl xyloccensin N were isolated from the fruit of the Chinese mangrove *Xylocarpus granatum*. Their structures were elucidated on the basis of modern spectroscopic techniques.

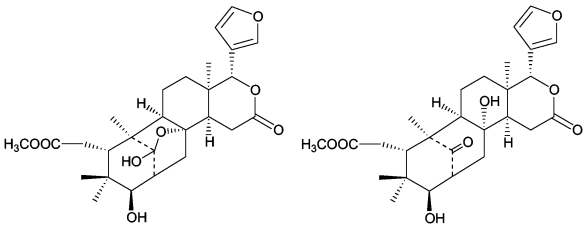
Key words: Mexicanolide, *Xylocarpus granatum*

Introduction

The family Meliaceae has proven to produce a variety of chemically unique antifeedant limonoids, such as azadirachtin [1 – 3] from the neem tree *Azadiracha indica* and harrisonin [4 – 5] from *Harrisonnia abyssinica*. Previous investigations on the seeds of two Meliaceae plants of mangrove, *X. granatum* and *X. moluccensis*, uncovered eleven limonoids, xyloccensins A – K [6 –,10]. In a continuing search for potential drug leads from Chinese tropical mangrove plants, we have recently reported the isolation and identification of three novel mexicanolides, named xyloccensins L – N [11 –,12], and eight unique 8, 9, 30-phragmalin ortho esters, named xyloccensins O – V [13 – 14] from the stem bark of *X. granatum*. Furthermore, chemical investigation of the fruit from the same plant resulted in the discovery of two new mexicanolides, named 3-deacetyl xyloccensin M and 3-deacetyl xyloccensin N. In this paper, we describe the isolation and structural elucidation of these compounds.

Results and Discussion

The ethanolic extract of the fruit of *X. granatum* was subjected to sequential extraction with petroleum ether and ethyl acetate as described in the Experimental section. The resulting ethyl acetate extract was chromatographed on silca gel, octadecylsilyl silica gel, Sephadex LH-20 gel and followed by preparative reverse-phase C_{18} HPLC to yield 3-deacetyl xyloccensin M (**1**) and 3-deacetyl xyloccensin N (**2**) (Fig. 1).



1 **2**

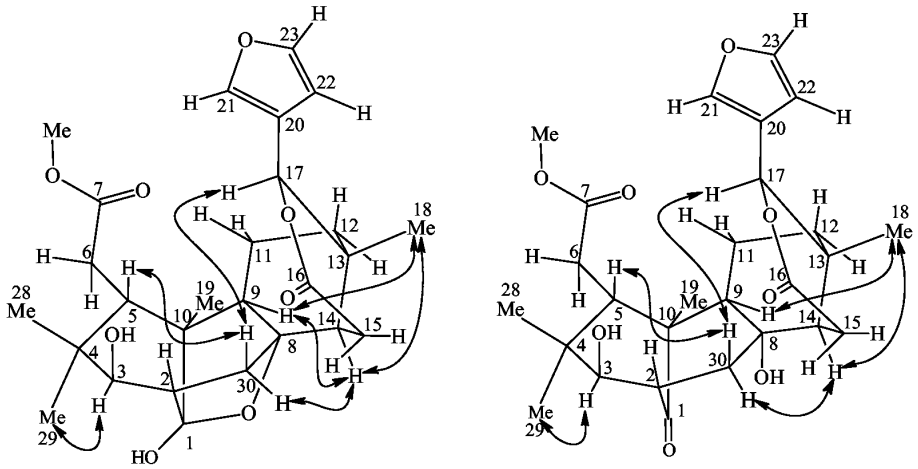
Fig. 1. Structures of compounds **1** and **2**.

The electrospary ionization (ESI)-MS (positive ion mode) of **1** showed pseudo-molecular peaks at *m/z* 511 $[M+Na]^+$ and 527 $[M+K]^+$, which proposed the molecular weight as 488. The HRESI-MS measurements indicated that the elemental composition was $C_{27}H_{36}O_8$, in agreement with the 1D NMR data (Table 1). Consequently, **1** had an unsaturation index of ten, which included two carbon-carbon double bonds, two ester functions and six rings. The ^1H and ^{13}C NMR spectral data (Table 1) of **1** were similar to those of xyloccensin M [11] except for the absence of a 3-acetyl group. In particular, the presence of an acetal carbon signal at $\delta = 109.8$ and an oxygenated quaternary carbon at $\delta = 81.0$ strongly suggested that **1** had the same ring structure as xyloccensin M [11]. Additionally, the significant NOE interactions (Fig. 2) observed from H-3 to Me-29, but not from H-3 to H-5, or from H-3 to H-30α helped to establish this 3β-hydroxyl group. Furthermore, NOE correlations (Fig. 2) from H-17 to H-30β and from H-30β to H-5 indicated a *cis* orienta-

0932–0776 / 05 / 1200–1291 $ 06.00 © 2005 Verlag der Zeitschrift für Naturforschung, Tübingen · http://znaturforsch.com

Table 1. ^{1}H and ^{13}C NMR data, HMBC and ^{1}H-^{1}H COSY correlations of deacetyl xyloccensin M (**1**) (500 and 125 MHz, methanol-d_4).

Carbon No.	^{1}H NMR (HMQC) δH; mult.; J(Hz)	^{13}C NMR δC; mult.	HMBC correlations	^{1}H-^{1}H COSY correlations
1		109.8; s		
2	2.70; d, 7.6	48.5; d	1, 3, 4, 30	3, 30α, 30β
3	3.74; d; 10.0	74.8; d	2, 4, 5, 29, 30	2
4		39.9; s		
5	2.77; br s	40.6; d	3, 4, 6, 7, 10, 19	6a, 6b
6	2.40; br s	33.6; t	4, 5, 7, 10	5
7		176.8; s		
8		81.0; s		
9	1.49; dd; 13.2; 2.0	64.7; d	5, 8, 10, 11	11α, 11β,
10		45.2; s		
11α	1.72; brdt; 13.2; 3.0	20.3; t	9, 13	9, 11β, 12α, 12β
11β	1.91; m		8, 9, 13	9, 11α, 12α, 12β
12α	1.35; m	35.0; t	9, 11, 17, 18	11α, 11β, 12β
12β	1.67; m		9, 11, 13, 17	11α, 11β, 12α
13		36.7; s		
14	2.12; d; 12.5	46.4 d	8, 9, 13, 15, 16, 17, 18	15α, 15β
15α	2.76; d; 4.5	28.9; t	8, 13, 14, 16	14, 15β
15β	2.92; d; 12.5		8, 13, 14, 16	14, 15α
16		173.4; s		
17	5.60; s	79.6; d	12, 13, 14, 16, 18, 20, 21, 22	
18	1.04; s	22.9; q	12, 13, 14, 17	
19	1.02; s	21.3; q	1, 5, 9, 10	
20		122.8; s		
21	7.65; br s	142.7; d	20, 22, 23	22
22	6.57; br s	111.0; d	20, 21, 23	23
23	7.54; br s	144.4; d	20, 21, 22	21, 22
28	1.14; s	23.0; q	3, 4, 5, 29	
29	0.82; s	25.1; q	3, 4, 5, 28	
30α	1.78; d; 13.2	29.8; t	2, 8, 9, 14	2, 30β
30β	2.52; dd; 13.2; 6.7		1, 2, 3, 4	2, 30α
7-OMe	3.74; s	52.3; q	7	



1 **2**

Fig. 2. Diagnostic NOE correlations in compounds **1** and **2**.

tion between these respective protons. Similarly, those (Fig. 2) from H-14 to Me-18 and H-9 also indicated their mutual *cis* relationship. Thus **1** was characterized as 3-deacetyl xyloccensin M.

Compound **2** was isolated as amorphous powder. Its molecular formula was established as $C_{27}H_{36}O_8$, which was the same as **1**, by HRESI-MS and NMR data (Table 2). The IR (3650 – 3200, 1740, 1708, 1635,

Table 2. ^{1}H and ^{13}C NMR data, HMBC and ^{1}H-^{1}H COSY correlations of deacetyl xyloccensin N (**2**) (500 and 125 MHz, methanol-d_4).

Carbon No.	^{1}H NMR (HMQC) δH; mult.; *J*(Hz)	^{13}C NMR δC; mult.	HMBC correlations	^{1}H-^{1}H COSY correlations
1		222.6; s		
2	3.07; m	50.0; d	1, 3, 4, 30	3, 30α, 30β
3	4.76; d; 8.7	78.1; d	2, 4, 5, 29, 30	2
4		41.5; s		
5	3.74; dd; 10.0; 2.5	44.1; d	3, 4, 6, 7, 10, 19	6a, 6b
6	2.45; br s	33.8; t	4, 5, 7, 10	5
7		176.2; s		
8		74.6; s		
9	1.78; dd; 13.2; 5.0	62.0; d	5, 8, 10, 11	11α, 11β
10		49.9; s		
11α	1.75; brdt, 13.0; 3.0	20.5; t	9, 12, 13	9, 11β, 12α, 12β
11β	2.41; m		8, 9, 12	9, 11α, 12α, 12β
12α	1.36; m	36.8; t	9, 11, 13, 17	11α, 11β, 12β
12β	1.70; m		9, 11, 17, 18	11α, 11β, 12α
13		37.0; s		
14	1.90; d; 4.5	52.1 d	8, 13, 15, 16, 17, 18, 30	15α, 15β
15α	2.88; d; 4.5	29.2; t	8, 13, 14, 16	14, 15β
15β	2.96; d; 8.0		8, 13, 14, 16	14, 15α
16		173.7; s		
17	5.91; s	79.4; d	12, 13, 14, 16, 18, 20, 21, 22	
18	1.04; s	23.5; q	12, 13, 14, 17	
19	1.09; s	19.4; q	1, 5, 9, 10	
20		122.5; s		
21	7.84; br s	143.0; d	20, 22, 23	22, 23
22	6.50; br s	111.1; d	20, 21, 23	21, 23
23	7.55; t; 1.5	144.4; d	20, 21, 22	21, 22
28	0.79; s	23.5; q	3, 4, 5, 29	
29	0.82; s	24.5; q	3, 4, 5, 28	
30α	2.02; d; 14.0	33.0; t	2, 8, 9, 14	2, 30β
30β	3.05; dd; 14.0; 7.0		1, 2, 3, 4	2, 30α
7-OMe	3.71; s	52.5; q	7	

875 cm^{-1}) data indicated the presence of carbon-carbon double bond, hydroxyl, keto and ester carbonyl groups. From the ^{1}H and ^{13}C NMR spectral data (Table 2), it was evident that five of ten unsaturation index were present as double bonds: two carbon-carbon double bonds (as furan ring) and three C=O (as one ketone and two esters). A β-furyl moiety and one methoxycarbonyl group were also apparent from the spectra. The ^{1}H and ^{13}C NMR spectral data (Table 2) of **2** were similar to those of xyloccensin N [11] except for the absence of an acetyl group in C-3. The significant NOE interactions observed from H-3 to Me-29, but not from H-3 to H-5, or from H-3 to H-30α helped to establish this 3β-hydroxyl group, which was the same as xyloccensin N. Additionally, the chemical shifts of C-8, C-9, C-14 ($\delta_C = 74.6, 62.0, 52.1$) of ring C was almost the same as those ($\delta_C = 73.7, 60.8, 51.9$) of xyloccensin N [11], suggested a α hydroxyl substituted at C-8 as that in xyloccensin N. And this was confirmed by the strong HMBC correlation from the proton of this hydroxyl to C-8. Furthermore, the significant NOE interactions (Fig. 2) observed from H-17 to H-30β also help to establish the α configuration of 8-OH (Fig. 2). Consequently, **2** was assigned as 3-deacetyl xyloccensin N.

Compounds **1** and **2** are a pair of isomers of mexicanolides. As viewed from the biosynthetic pathway, **2** may be the possible biosynthetic intermediate of **1**. And it represented to our knowledge that this was the second time to get a pair of isomers of mexicanolides from the plant simultaneously.

Experimental Section

General

NMR spectra were recorded in methanol-d_4 using a Bruker AV-500 spectrometer (500 MHz for ^{1}H NMR and 125 MHz for ^{13}C NMR) with tetramethylsilane as the internal standard. Electrospary ionization (ESI)-MS spectra were measured on a Bruker APEX II spectrometer in positive ion mode. Optical rotations were recorded on a POLAPTRONIC HNQW5 automatic high-resolution polarimeter (Schmidt & Haensch Co. Ltd.) Preparative HPLC was carried out on ODS columns (250 × 10 mm i.d., YMC) with a Waters 996 photodiode array detector. For CC, silica gel (200 – 300 mesh) (Qingdao Mar. Chem. Ind. Co. Ltd.), oc-

tadecylsilyl silica gel (80 – 100 μm) (Unicorn) and Sephadex LH-20 gel (Pharmacia) were used.

Plant material

The fruit of *Xylocarpus granatum* was collected in October 2004 from Hainan Island, southern China. The identification of the plant was performed by Prof. Yongshui Lin, Laboratory of Marine Biology, South China Sea Institute of Oceanology, Chinese Academy of Sciences. A voucher sample (NO. GKLMMM-002-2) is kept in the Herbarium of South China Sea Institute of Oceanology.

Extraction and isolation

The dried fruit (4.5 kg) of *X. granatum* was extracted with hot 95% ethanol three times respectively. After removal of the solvent by evaporation, the residue was suspended in water and defatted with petroleum ether. The aqueous layer was further extracted with ethyl acetate. The resulting ethyl acetate extract (105 g) was subjected to silica gel CC using chloroform-methanol system (100:0 $\sim$ 2:1) to yield 70 fractions. Fractions 10 to 16 (4 g) were combined and subjected to CC on silica gel, octadecylsilyl silica gel and Sephadex LH-20 gel, followed by preparative reverse-phase C_{18} HPLC using acetonitrile-water system (30:70) to yield compounds **1** (12 mg) and **2** (8 mg).

*Deacetyl xyloccensin M (**1**)*

Amorphous powder, $[\alpha]_D^{25}$ -70° (c 0.6, methanol). – IR (KBr): $\tilde{\nu}$ = 3450, 3140, 1730, 1635, 870 cm^{-1}. – ^{1}H NMR (500 MHz, methanol-d_4) and ^{13}C NMR (125 MHz, methanol-d_4): See Table 1. – MS (HR-ESI): $m/z = 511.2306$ [M+Na]$^+$. ($C_{27}H_{36}O_8Na$ requires 511.2308)

*Deacetyl xyloccensin N (**2**)*

Amorphous powder, $[\alpha]_D^{25}$ $+15^\circ$ (c 0.8, methanol). – IR (KBr): $\tilde{\nu}$ = 3650 – 3200, 1740, 1708, 1635, 875 cm^{-1}. – ^{1}H NMR and ^{13}C NMR (methanol-d_4): See Table 1; – HR-ESI-MS, $m/z = 511.2310$ [M+Na]$^+$. ($C_{27}H_{36}O_8Na$ requires 511.2308)

Acknowledgements

Support for this work from the Important Project of Chinese Academy of Sciences (KZCX3-SW-216) and the Natural Science Foundation of Guangdong Province (04100729) is gratefully acknowledged. Mass spectra were provided by Institute of Chemistry, Chinese Academy of Sciences and the NMR spectra by the Laboratory of NMR Analysis and Measurement, South China Sea Institute of Oceanology, Chinese Academy of Sciences.

[1] D. E. Champagne, O. Koul, M. B. Isman, G. G. E. Scudder, G. H. N. Towers, Phytochemistry **31**, 377 (1992).
[2] J. H. Butterworth, E. D. Morgan, Chem. Commun. 23 (1968).
[3] D. A. H. Taylor, Tetrahedron **43**, 2779 (1987).
[4] M. S. Rajab, J. K. Rugutt, F. R. Fronczek, N. H. Fischer, J. Nat. Prod. **60**, 822 (1997).
[5] I. Kubo, S. P. Tanis, Y. W. Lee, I. Miura, K. Nakanishi, A. Chapya, Heterocycles **5**, 485 (1976).
[6] A. S. Ng, A. G. Fallis, Can. J. Chem. **57**, 3088 (1979).
[7] I. Kubo, I. Miura, K. Nakanishi, J. Am. Chem. Soc. **98**, 6704 (1976).
[8] K. A. Alvi, P. Crews, B. Aalbersberg, R. Prasad, J. Simpson, R. T. Weavers, Tetrahedron **47**, 8943 (1991).
[9] D. A. Mulholland, B. Parel, P. H. Coombes, Curr. Org. Chem. **4**, 1011 (2000).
[10] U. Kokpol, W. Chavasiri, S. Tip-pyang, G. Veerachato, F. L. Zhao, Phytochemistry **41**, 903 (1996).
[11] J. Wu, S. Zhang, Q. Xiao, Q. X. Li, J. S. Huang, Z. H. Xiao, L. J. Long, Z. Naturforsch. **58b**, 1216 (2003).
[12] J. Wu, S. Zhang, Q. Xiao, Q. X. Li, J. S. Huang, L. J. Long, L. M. Huang, Tetrahedron Lett. **45**, 591 (2004).
[13] J. Wu, Q. Xiao, J. S. Huang, Z. H. Xiao, S. H. Qi, Q. X. Li, S. Zhang, Org. Lett. **6**, 1841 (2004).
[14] J. Wu, Q. Xiao, S. Zhang, X. Li, Z. H. Xiao, H. X. Ding, Q. X. Li, Tetrahedron **61**, 8382 (2005).