Studies on chemical constituents of planted *Taxus mairei* (Ⅲ)

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Abstract: From the chloroform-soluble fraction of the ethanol extracts of the whole plant of *Taxus chinensis* var. *mairei* (Lemee et Levl), four compounds were isolated by using repeated column chromatography on silica gel and Sephadex LH-20. Based on spectroscopic data (UV, IR, ESI-MS, ^{1}H NMR and ^{13}C NMR), the compounds were identified as taxamairin K (**1**), 2α, 4α-dideacetoxy-7β-benzoyloxy-5β,20-epoxy-9α,10β,13α,15-tetrahydroxy-11(15→1)abeotaxa-11-ene (**2**), 7β-xylosyl-taxol (**3**), 10-deacetoxy-7-xylosyl-taxol (**4**). Among them, taxamairin K is a new compound.

Key words: *Taxus chinensis* var *mairei* (Lemee et Levl); taxamairin K; planted

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人工栽培的南方红豆杉化学成分研究(Ⅲ)

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摘要: 从人工栽培的南方红豆杉全株乙醇提取物的氯仿萃取部位通过反复硅胶和凝胶柱层析分离得到4个化合物,采用波谱解析(IR,ESI-MS,^{1}H NMR和^{13}C NMR)等方法确定了其结构,4个化合物分别鉴定为 taxamairin K (**1**), 2α,4α-dideacetoxy-7β-benzoyloxy-5β,20-epoxy-9α,10β,13α,15-tetrahydroxy-11(15→1)abeotaxa-11-ene (**2**), 7β-xylosyl-taxol (**3**), 10-deacetoxy-7-xylosyl-taxol (**4**),其中化合物**1**为新化合物。

关键词: 南方红豆杉; taxamairin K; 人工栽培

Taxol, a highly effective anticancer agent, couldn't meet the need of clinical use because of its scare source. There are many ways to solve this shortage, such as planting, synthesis, and semisynthesis, tissue and cell culture and biotranformation. Planting and semisynthesis are most viable for the difficulty. *Taxus chinensis* var. *mairei* grow mainly in Fujian, Taiwan and Jiangxi Province[1]. After doing research for getting planted species for several years, we succeed in planting widely the species. The content of taxol in planted *Taxus mairei* is about 0.01% nearly to those of the wild species[2,3]. To study whether the chemical constituents of planted *Taxus mairei* are the same as those of the wild ones, we investigated the whole tree of *T. chinensisi* var. *mairei* planted in Longyan, Fujian Province[4,5]. From the chloroform extracts, a new taxamairin and three known compounds were obtained named as taxamairin K (**1**), 2α,4α-dideacetoxy-7β-benzoyloxy-5β,20-epoxy-9α,10β,13α,15-tetrahydroxy-11(15→1)abeotaxa-11-ene (**2**)[6], 7β-xylosyl-taxol (**3**)[7], 10-deacetoxy-7-xylosyl-taxol (**4**)[8].

Result and discussion

Compound **1** was obtained as yellow crystals. The molecular formula was established as $C_{19}H_{18}O_6$ by HR-ESI-MS (*m/z* 343.152 0 [M + H]$^+$, calcd for 343.154 0) and negative ESI-MS (*m/z* 341.1 [M - H]$^-$), which could be supported by the data of

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^{1}H NMR and ^{13}C NMR spectra. The IR spectrum was attributable to the existence of carbonyl groups (1 690 cm^{-1}, 1 753 cm^{-1}). The ^{1}H NMR and ^{13}C NMR spectra showed existence of four unsaturated hydrogen [δ 7.31(1H,d,J=9.48), 6.20(1H,d,J=9.48), 6.82(1H,s), 7.91(1H,s)], four methyl groups [δ 1.49(6H,s), 1.32(6H,d,J=6.93), (δ 27.3, 20.0)], two methenyl groups [(δ 6.82(1H,s), 2.86(1H,q,J=6.87), (δ 99.8, 33.3)]. The ^{13}C NMR spectrum exhibited signals of three carbonyl groups (δ 184.3, 199.7, 171.6). The signal 1 690 cm^{-1} of IR, the signal of δ 199.7 in ^{13}C NMR, and the AB double signal of δ 7.31(1H,d,J=9.48), 6.20 (1H,d,J=9.48) suggested the existence of an α,β-unsaturated ketone, supported by the HMBC spectrum (the correlations of δ 7.31(1H,d,J=9.48) and δ 199.7). The signal 1 753 cm^{-1} of IR and the signal of δ 171.6 in ^{13}C NMR suggested the existence of an ester function. Some spectra characters of compound **1** are the same as those of taxamairin A[9], especially in ring A and ring B. In the HMBC spectrum, the correlations of H-15 (δ 6.82,1H,s) with the C-16 (δ 171.6), C-1 (δ 184.3) and C-3 (δ 121.4) indicated that C-15 connect with C-2 (δ 148.3). The cross-peaks of H-4 (δ 7.91) with C-14 (δ 163.4) in the HMBC spectrum, revealed that C-14 connect to C-3. The structure of ring C was determined by above data. In the HMBC spectrum, the correlations of H-17 (δ 2.86,1H,q,J=6.87) with the carbonyl group δ 171.6(C-16) and the methyl group δ 20.0(C-18, 19) determined the chain structure of ring C. Thus, the structure of the new compound was determined completely, named as taxamairin K (Figure 1).

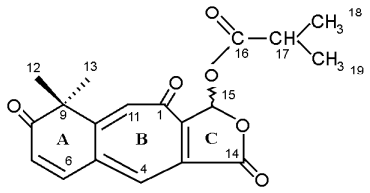

Figure 1 The structure of compound **1**

Experimental

1 General experimental procedures

Melting points were obtained by XT-4 microthermoanalysis apparatus (uncorrected). Optical rotations were determined on PE-241MC polarimeter. IR spectra were recorded on a Perkin-Elemer 983 spectrophotometer with KBr pellets. NMR experiments were performed on a Bruker ACF-300 and an ACF-500 spectrometer, TMS was used as the internal standard. MS was recorded on a VG Auto Spec-3000 spectrometer. Silica plate for TLC and silica gel (200 - 300 mesh) for column chromatography were obtained from Qingdao Marine Chemical Corporation (Qingdao, China).

Table 1 NMR data for compound **1** (300 MHz for ^{1}H and 75 MHz for ^{13}C in $CDCl_3$)

Position	^{1}H NMR δ	^{13}C NMR δ	HMBC(H→C)
1		184.3	
2		148.3	
3		121.4	
4	7.91(1H,s)	134.7	2,3,5,6,10,14
5		135.0	
6	7.31(1H,d,J=9.48)	146.4	5,8,10
7	6.20(1H,d,J=9.48)	125.6	5,9
8		199.7	
9		50.7	
10		154.8	
11	7.15(1H,s)	134.4	1,2,5,9,10
12	1.49(3H,s)	27.3	8,9,10,13
13	1.49(3H,s)	27.3	8,9,10,12
14		163.4	
15	6.82(1H,s)	99.8	1,3,16,17
16		171.6	
17	2.86(1H,q,J=6.87)	33.3	16,18,19
18	1.32(3H,d,J=6.93)	20.0	16,17,19
19	1.32(3H,d,J=6.93)	20.0	16,17,18

2 Plant material

Taxus chinensis var. *mairei* (Lemee et Levl) was collected in Longyan, Fujian Province and identified by engineer Li Wenjian (Fujian South Biotechnology Co. Ltd.).

3 Extraction and isolation

The dried whole plant (20 kg) of *T. chinensis* var. *mairei* was powdered and extracted with 75% EtOH. After evaporation to dryness under reduced pressure, the residue was suspended in water and then extracted successively with petroleum ether and chloroform. The chloroform-soluble part (160 g) was chromatographed on silica gel ($CHCl_3$-CH_3OH 100:0→100:10) to give seven fractions, which were subjected to further separation using repeated silica gel and Sephadex LH-20 column chromatography, to yield **1** (50 mg, from fraction 1), **2** (20 mg, from fraction 3), **3** (120 mg, from fraction 5), **4** (1 g, from fraction 7).

4 Identification

Compound 1 Yellow crystal (chloroform / petroleum ether), mp 165 – 168 ℃; $[\alpha]_D^{20}$ – 13.2 (*c* 0.06, MeOH); HR-ESI-MS *m/z*: 343.152 0 [M + H]$^+$, calcd for 343.154 0; ESI-MS *m/z*: 341 [M – H]$^-$; UV λ_{max}^{MeOH} nm (logε): 409.1 (0.4), 269.8 (0.2); IR_{max}^{KBr} cm^{-1}: 3 020, 2 999, 2 970, 2 870, 2 698, 2 667, 2 586, 2 546, 1 753, 1 690, 1 605, 1 456, 1 418, 1 371, 1 306, 1 221, 1 186, 916 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) and ^{13}C NMR (75 MHz, $CDCl_3$) δ see Table 1.

References

[1] Liang JY, Huang KS. Progress in Medicinal Chemistry: Vol. 9 (药物化学进展:第一卷) [M]. Beijing: Chinese medical Technology Press, 1999:96 – 115.

[2] Su YJ, Wang T, Li XY, et al. Analysis on the amounts of taxol in different location of *Taxus chinensis* var. *Mairei* [J]. Nat Prod Res Dev (天然产物研究与开发), 2001,13:19 – 20.

[3] Zheng DY. Study on the taxol content in different parts of Taxus growing in China [J]. J Fujian Coll For (福建林学院学报), 2003,23:160 – 163.

[4] Chen XY, Liang JY. Studies on chemical constituents of planted *Taxus mairei* (I) [J]. Chin J Nat Med (中国天然药物), 2006,4:52 – 57.

[5] Chen XY, Liang JY. Studies on chemical constituents of planted *Taxus mairei* (II) [J]. Chin J Nat Med (中国天然药物), 2006,4:101 – 103.

[6] Shi QW, Oritani T, Sugiyama T, et al. Three new rearranged taxane diterpenoids from the bark of *Taxus chinensis* var. *mairei* and the needles of *Taxus cuspidata* [J]. Heterocycles, 1999,51:841 – 850.

[7] Kingston DGI. The Taxane Diterpenoids Progress in the Chemistry of Organic Natural Products: Vol 61 [M]. New York: Spring-Verlog, 1993:1 – 206.

[8] Pao GH. Study on chemical consitituents of root of *Taxus yunnanensis* [D]. Nanjing: China Pharmaceutical University, 1996.

[9] Liang JY, Min ZD, Tanaka T, et al. Studies on the diterpenes from *Taxus mairei* I. Structures of taxamairins A, B and C [J]. Acta Chim Sin (化学学报), 1988, 46:21 – 25.