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Synthesis and Biological Evaluation of 4-Deacetoxy-1,7-dideoxy Azetidine Paclitaxel Analogues

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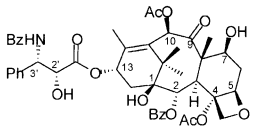
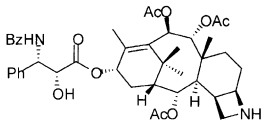
Abstract—Three novel 4-deacetoxy-1,7-dideoxy azetidine paclitaxel analogues were synthesized through a convenient route that employed hydroboration-amination and intramolecular S_N2-type substitution reaction from a natural taxoid taxinine. All analogues have been tested for cytotoxicity against three human tumor cell lines. None of them showed remarkable cytotoxicity compared to paclitaxel against tested tumor cell lines.



paclitaxel (1)



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Paclitaxel[1] (**1**, Taxol®) and docetaxol[2] (Taxotere®) are clinically used in the treatment of advanced ovarian and breast cancers. Both also exhibit potent antitumor activity against various cancer cells which have been uneffectively treated by existing chemotherapeutic drugs.[3] Paclitaxel has also been approved for the second-line treatment of AIDS related Kaposi's sarcoma in 1997.[4] Since its discovery at the end of 1960s, a number of chemical, biological and medicinal researches as well as the structure–activity relationships (SAR) of paclitaxel have been carried out, especially in the recent decade.[5] Therefore, several major problems have been emerged such as low aqueous solubility, multi-drug resistance (MDR), scarcity of the drug and little activity for oral administration. To overcome these problems, searching for novel promising paclitaxel analogues as anticancer drug candidates is still necessary. SAR studies so far indicated that the oxgen functions at 7, 9, 10-positions were not essential for paclitaxel's activity,[6] while the 13-side chain and 2-benzoyloxy group were crucial[7] (He et al.[8] reported that *m*-azido group at 2-benzoyloxy group could be a substitute for the 13-side chain, on the contrary to our results[9]). Although the necessity of 1-OH group is still unclear,[10] the presence of oxetane ring and 4-acyloxy group[11] was shown to be essential: azetidine type[12] or 4-deacetoxyl analogues lost the activity.[13] On our continuing efforts to evaluate SAR of paclitaxel,[9,14] we planned to prepare novel 4-deacetoxy paclitaxel analogues bearing azetidine ring. Here we report a convenient synthesis and antitumor activity of some 4-deacetoxy-1,7-dideoxy azetidine paclitaxel analogues, such as **2**, from naturally abundant taxinine (**3**).

Synthetic protocol to 1-deoxypaclitaxel azetidine analogues included the synthesis of the known taxane **4**[15] which could be prepared through 2 steps from naturally abundant taxinine **3** isolated from the Japanese yew, *Taxus cuspidata* (2 g/kg from the bark). As the synthetic protocol depicted in Scheme 1, **4** was converted to bis-*O*-silyl compound **5**. The double bond at C-4(20) was subjected to hydroboration-amination to form primary amine **6** in 86% yield.[16] Protection of the amino group and selective deprotection of the TES group gave alcohol **7**. Mesylation of the resulting hydroxyl group of **7** followed by alkaline treatment yielded a key intermediate azetidine taxane **8**.[12,17] Deprotection of the 13-TBS group and subsequent esterification with

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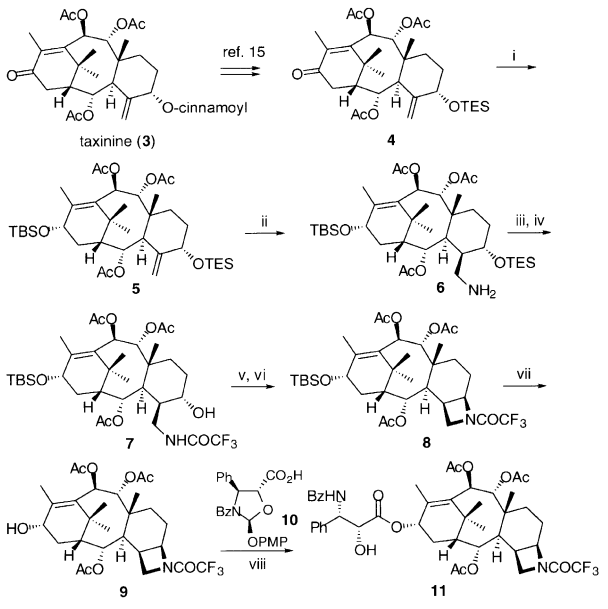
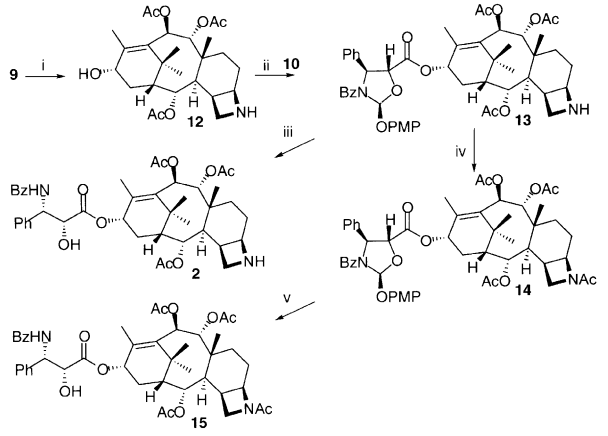


Scheme 1. Reagents and conditions: (i) (a) $NaBH_4$, $CeCl_3$, MeOH, rt, 73%; (b) TBSCl, imidazole, DMF, rt, 91%. (ii) (a) BH_3•THF, THF, rt, (b) NH_2OH, NaOCl, rt, 86%; (iii) $(CF_3CO)_2O$, DMAP, Py, rt, 82%; (iv) AcOH–H_2O–THF (4:3:11), rt, 12 h, 84%; (v) MsCl, pyridine, CH_2Cl_2, rt, 20 h, 83%; (vi) DMSO, K_2CO_3, rt, 56%; (vii) TBAF, THF, rt, 98%; (viii) (a) **10**, DCC, DMAP, toluene, 70 °C; (b) TFA–H_2O, 65% two steps.

(2*R*,4*S*,5*R*)-3-benzoyl-2-(*p*-methoxyphenyl)oxy-4-phenyl-1,3-oxazolidine-5-carboxylic acid (**10**) followed by acid hydrolysis afforded the expected 4-deacetoxy-1,7-dideoxy paclitaxel azetidine compound **11** according to the previously established methods.[14c–d]

As shown in Scheme 2, the compound **9** was reduced using $NaBH_4$ to give a free azetidine **12** in 66% yield. Esterification of the azetidine **12** with the carboxylic acid **10** provided ester **13**, which was hydrolyzed to afford 4-deacetoxy-1,7-dideoxy paclitaxel azetidine analogue **2** in 88% yield. Additionally, *N*-acetyl derivatives of the azetidine ring, compounds **14** and **15**, were prepared in the similar manner.



Scheme 2. Reagents and conditions: (i) $NaBH_4$, EtOH, rt, 66%; (ii) **10**, DCC, DMAP, toluene, 70 °C, 73%; (iii) TFA–H_2O, rt, 88%; (iv) Ac_2O, DMAP, rt, 12 h, 85%; (v) TFA–H_2O, rt, 81%.

Table 1. Results of biological assays in vitro on the cytotoxicity of compounds **2**, **11**, **13**, **14** and **15**

Compd	Cytotoxicity against tumor cell (ED_{50}/ED_{50})[a,b,c]		
	SK-OV3	MCF-7	A549
Paclitaxel (**1**)	1.0	1.0	1.0
2	**4.6**	**4.9**	**4.5**
11	**4.7**	**3.8**	**3.2**
13	9.3	8.6	6.8
14	10.2	9.3	6.9
15	**5.8**	**3.6**	**2.9**

[a]ED_{50} is the concentration of 50% inhibition of proliferation after 72 h of incubation.
[b]Ratio of ED_{50} relative to paclitaxel is 1.0.
[c]SK-OV3: ovarian cancer cell lines, MCF-7: breast cancer cell lines, A549: lung cancer cell lines.

All new 4-deacetoxy-1,7-dideoxy paclitaxel azetidine analogues **2**, **11**, **13**, **14** and **15** were evaluated in vitro on the cytotoxicity against three tumor cell lines: SK-OV3 (ovarian cancer), MCF-7 (breast cancer) and A549 (lung cancer). As presented in Table 1, compared to paclitaxel (**1**), all the tested compounds showed a decrease of cytotoxicity against three tumor cell lines. This demonstrates that the 4-acetoxy group and the oxetane ring are essential for activity. However, compared to the former azetidine analogue **12** and the 4-deacetoxy analogue **13**, our analogues with the original 13-side chain (**2**, **11** and **15**) still retained some activity. We think that the azetidine NH group could compensate the polarity of the original circumstances of paclitaxel. In addition, the results also support the hypothesis that 1-OH group is not essential for the activity.

Acknowledgements

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References and Notes

1. Wani, M. C.; Taylor, H. L.; Wall, M. E.; Coggon, P.; McPhail, A. T. *J. Am. Chem. Soc.* **1971**, *93*, 2325.
2. Guenard, D.; Gueritte-Vogelein, F.; Potier, P. *Acc. Chem. Res.* **1993**, *26*, 160.
3. (a) Bissery, M. C.; Guenard, D.; Gueritte-Vogelein, F.; Lavelle, F. *Cancer Res.* **1991**, *51*, 4845. (b) Schiff, P. B.; Fant, J.; Horwitz, S. B. *Nature* **1979**, *227*, 665.
4. Ojima, I.; Wang, T.; Miller, M. L.; Lin, S.; Borella, C. P.; Geng, X.; Pera, P.; Bernacki, R. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 3423.
5. (a) Gueritte-Vogelein, F.; Guenard, D.; Dubois, J.; Wahl, A.; Marder, R.; Muller, R.; Lund, M.; Bricard, L.; Potier, P. In *Taxane Anticancer Agents: Basic Science and Current Status*, Georg, G. I., Chen, T. T., Ojima, I., Vyas, D. M., Eds.; ACS Symposium Series 583; American Chemistry Society: Washington, DC, 1995; pp 189–202. (b) Kingston, D. G. I. In *Taxane Anticancer Agents: Basic Science and Current Status*, Georg, G. I.; Chen, T. T.; Ojima, I.; Vyas, D. M., Eds.; ACS Symposium Series 583, America Chemistry Society: Washington, DC, 1995; p 203.

6. Chaudhary, A. G.; Rimoldi, J. M.; Kingston, D. G. I. *J. Org. Chem.* **1993**, *58*, 3798.
7. Nicolaou, K. C.; Renaud, J.; Nantermet, P. G.; Couladouros, E. A.; Guy, R. K.; Arasidlo, W. *J. Am. Chem. Soc.* **1995**, *117*, 2409.
8. He, L.; Jagtap, P. G.; Kingston, D. G. I.; Scen, H. J.; Orr, G. A.; Horwitz, S. B. *Biochemistry* **2000**, *39*, 3972.
9. Horiguchi, T.; Kiyota, H.; Oritani, T. *Tetrahedron*, in press.
10. Kingston, D. G. I.; Chordia, M. D.; Jagtap, P. G. *J. Org. Chem.* **1999**, *64*, 1814.
11. Chen, S. H.; Wei, J. M.; Long, B. H.; Fairchild, C. A.; Carboni, J.; Member, S. W.; Rose, W. C.; Johnoston, K.; Casazza, A. M.; Kadow, J. F.; Farina, V.; Vyas, D.; Doyle, T. W. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 2741.
12. Marder-Karsenti, R.; Dubois, J.; Bricard, L.; Guenard, D.; Gueritte-Voegelein, F. *J. Org. Chem.* **1997**, *62*, 6631.
13. (a) Magri, N. F.; Kingston, D. G. I. *J. Org. Chem.* **1986**, *51*, 797. (b) Chordia, M. D.; Chaudhary, A. G.; Kingston, D. G. I.; Jiang, Y. Q.; Hamel, E. *Tetrahedron Lett.* **1994**, *35*, 6843.
14. (a) Cheng, Q.; Oritani, T.; Horiguchi, T. *Tetrahedron* **1999**, *55*, 12099. (b) Cheng, Q.; Oritani, T.; Horiguchi, T.; Yamada, T.; Hassner, A. *Tetrahedron* **2000**, *56*, 1181. (c) Cheng, Q.; Oritani, T.; Horiguchi, T.; Yamada, T.; Meng, Y. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 517. (d) Cheng, Q.; Oritani, T.; Horiguchi, T. *Tetrahedron* **2000**, *56*, 1667. (e) Horiguchi, T.; Cheng, Q.; Oritani, T. *Tetrahedron Lett.* **2000**, *41*, 3907. (f) Horiguchi, T.; Nagura, M.; Cheng, Q.; Oritani, T.; Kudo, T. *Heterocycles* **2000**, *53*, 2629. (g) Cheng, Q.; Horiguchi, T.; Yamaguchi, M.; Oritani, T. *Heterocycles* **2000**, *53*, 2759.
15. Hosoyama, H.; Shigemori, H.; In, Y.; Ishida, T.; Kobayashi, J. *Tetrahedron* **1998**, *54*, 2521.
16. Preparation procedure: to a solution of taxoid **5** (57 mg, 0.10 mmol) in THF (8 mL) was slowly injected BH_3·THF complex (0.2 mL, 1.0 M in THF) under nitrogen at 0 °C. The mixture was stirred for an additional 2 h at 0 °C and then allowed to warm rt, and methanol (0.2 mL) was added. After removal of THF, the residue was resolved in CH_2Cl_2 (5 mL) followed by the addition of NH_2OH·HCl (34 mg, 0.50 mmol, 5 equiv) and NaOCl (2 mL). The resulting mixture was stirred at rt for 12 h and then extracted with EtOAc. The organic phase was washed with water and brine, and dried over $MgSO_4$. After removal of the solvent, the residue was purified on silica gel (EtOAc/hexane = 2/1) to give **6** (51 mg, 86%).
17. Fenoglio, F.; Nano, G. M.; Vander Velde, D. G.; Appendino, G. *Tetrahedron Lett.* **1996**, *37*, 3203.