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Synthesis of (−)-4-aza-4-deoxypodophyllotoxin from (−)-podophyllotoxin

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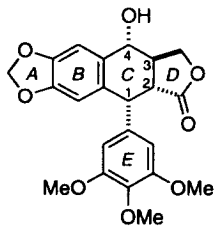
Abstract

(−)-4-Aza-4-deoxypodophyllotoxin (**4**) was synthesized from (−)-podophyllotoxin (**1**) through C-ring cleavage, Curtius rearrangement and intramolecular *N*-alkylation. Analogue **4** showed potent cytotoxicity against P388 leukemia cells.

Keywords: podophyllotoxin; 4-aza-4-deoxypodophyllotoxin; Curtius rearrangement; cytotoxicity.

Microtubules are an important target in the development of novel anticancer drugs, and *Catharanthus* alkaloids, vinblastine and vincristine, and taxol have already been used clinically. Podophyllotoxin (**1**) is also known to bind to tubulin, but its binding site differs from that of vinblastine[1] and taxol.[2] This evidence suggests that podophyllotoxin (**1**) may become a lead compound having activity against tumors resistant to the above drugs. Recently, podophyllotoxin heterocyclic analogues have attracted much attention since some of them showed more promising antitumor activity and were less toxic than podophyllotoxin (**1**).[3] The findings that 2,4-diaza-4-deoxypodophyllotoxin (**2**) showed significant antitumor activity against vincristine-resistant P388 leukemia (P388/VCR) cells in vivo,[4] while podophyllotoxin (**1**)[4] and 2-aza-4-deoxypodophyllotoxin (**3**)[5] showed only marginal or weak activity, suggested that substitution of the carbon atom at the position 4 by a nitrogen atom would be important to bring about such changes in the biological profile. We describe herein the synthesis of (−)-4-aza-4-deoxypodophyllotoxin (**4**) which has not been known thus far.



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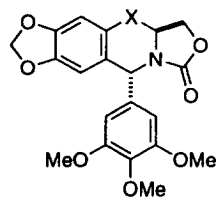
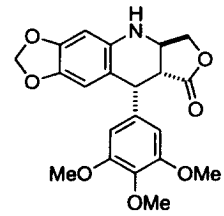


2: X = NH
3: X = CH_2



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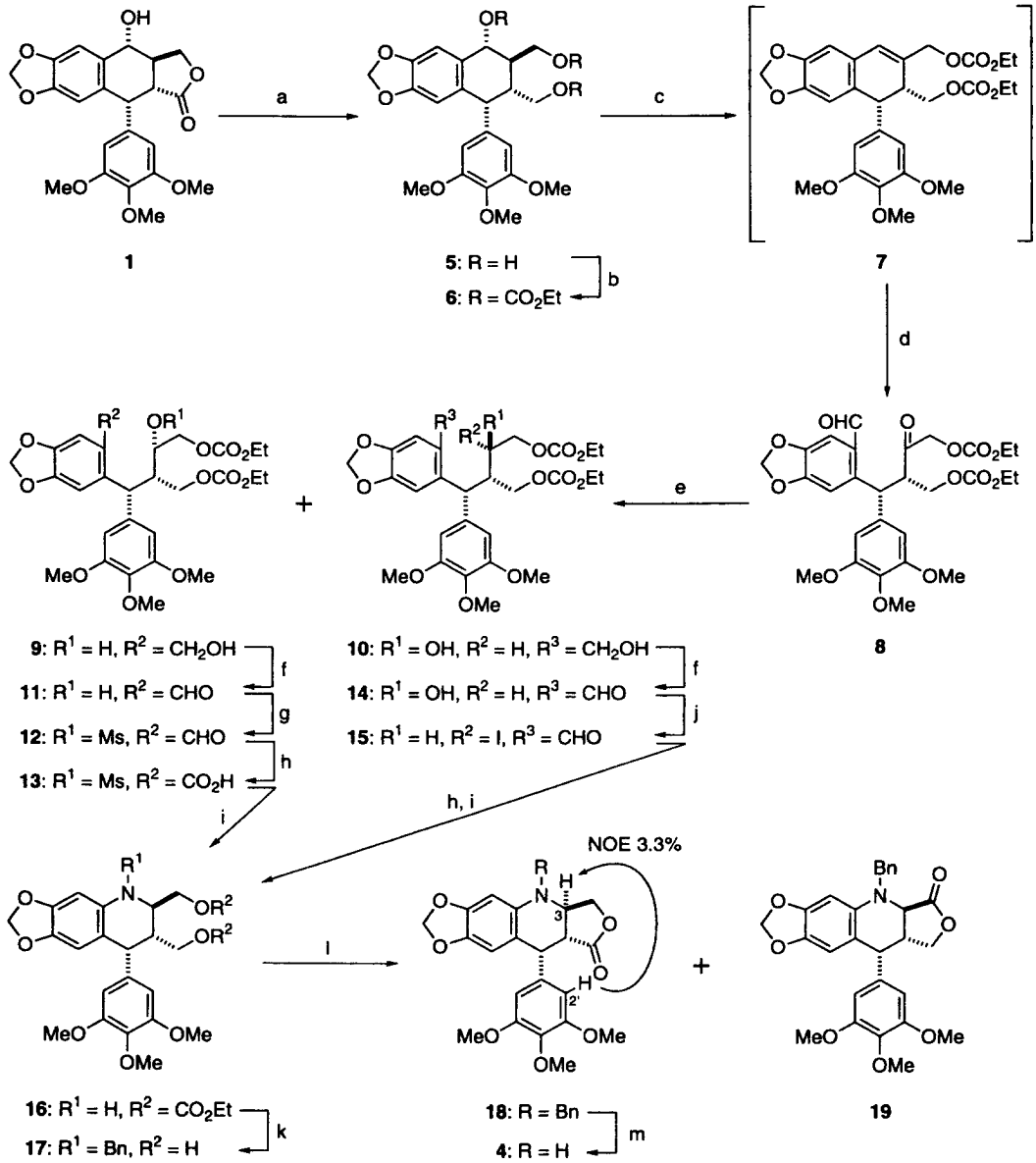
Since the configuration at the C-1 center is known to be critical for expressing the activity,[6] we planned to synthesize this compound in an optically pure form from natural (–)-podophyllotoxin (**1**). To avoid loss in chiral integrity at the C-2 position throughout the transformation, which would occur readily under basic conditions, we reduced the lactone D-ring in the initial step. Lithium aluminum hydride reduction of **1** gave triol **5** in 71% yield (Scheme 1).[7] Compound **5** was then treated with ethyl chloroformate in pyridine to afford tricarbonate **6** quantitatively. Interestingly, when compound **6** was heated in 1-methylnaphthalene at 170°C for 1.75 h, the ethoxycarbonyloxy group at the position



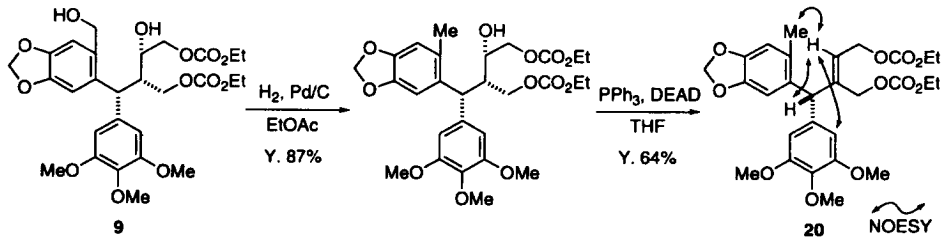
Scheme 1. *Reagents and conditions*: (a) $LiAlH_4$, THF, room temp., 71%; (b) $ClCO_2Et$, pyridine, –20°C, quant.; (c) 1-methylnaphthalene, 170°C; (d) O_3, MeOH–$CHCl_3$, –78°C, then Me_2S, 56% from **6**; (e) BH_3, THF, room temp., 24% (**9**) and 63% (**10**); (f) MnO_2, CH_3CN, room temp., 84% for **11**, 85% for **14**; (g) MsCl, Et_3N, $CHCl_3$, room temp., 84%; (h) $NaClO_2$, 2-methyl-2-butene, *t*-BuOH, pH 4 buffer, quant. for **13**; (i) DPPA, Et_3N, dioxane, room temp. ~120°C, 69% from **13**, 34% from **15**; (j) PPh_3, I_2, imidazole, PhMe, 90°C, 68%; (k) BnBr, K_2CO_3, MeOH–H_2O, 65°C, 82%; (l) 4-methylmorpholine *N*-oxide, *n*-Pr_4NRuO_4, MS 4A, CH_2Cl_2, room temp., 26% (**18**) and 33% (**19**); (m) H_2, Pd/C, AcOH–EtOH, 89%

4 was selectively eliminated to give a rather unstable dihydronaphthalene intermediate **7**, which was then subjected to ozonolysis to produce ketoaldehyde **8** in 56% yield from **6**. Reduction of **8** using a borane–tetrahydrofuran complex gave epimeric diols **9** and **10** in yields of 24 and 63%, respectively.[8,9] Diols **9** and **10** were converted into the same intermediate **16** separately. Selective oxidation of diol **9** using activated manganese dioxide gave aldehyde **11** in 84% yield, which was then converted to mesylate **12** in 84% yield. Chlorite oxidation of **12** gave carboxylic acid **13** quantitatively.[10] Curtius rearrangement of **13** using diphenylphosphoryl azide (DPPA), and successive intramolecular *N*-alkylation of the resultant amine intermediate produced tetrahydroquinoline **16** in 69% yield. The epimeric diol **10** was oxidized to aldehyde **14** in 85% yield, and then converted into iodide **15** in 68% yield by using triphenylphosphine, iodine and imidazole.[11] Chlorite oxidation of **15** gave an unstable carboxylic acid, which was then subjected to Curtius rearrangement to provide **16** in 34% yield from **15**.[12] Treatment of **16** with benzyl bromide under basic aqueous conditions resulted in *N*-benzylation and decarbonylation to afford diol **17** in 82% yield. Catalytic oxidation of **17** using tetrapropylammonium perruthenate, 4-methylmorpholine *N*-oxide and powdered molecular sieves 4A provided lactones **18** and **19** in yields of 26 and 33%, respectively.[13] Debenzylation of **18** gave compound **4** in 89% yield. The stereochemistry of this compound was confirmed by the ^{1}H NMR spectrum observing 3.3% nuclear Overhauser enhancement between H-2′ and H-3, and a large coupling constant of 12.8 Hz between H-2 and H-3 having an antiperiplanar relationship.[14]

A preliminary biological evaluation of analogue **4** and compound **1** was performed using P388 leukemia cells. Both compounds showed the same IC_{50} value of 0.0050 μg/mL. The in vivo antitumor activity of analogue **4** against P388/VCR cells will be reported in due course.[15]

References

1. Wilson, L. *Biochemistry* **1970**, *9*, 4999–5007.
2. Parness, J.; Horwitz, S. B. *J. Cell Biol.* **1981**, *91*, 479–487; Vallee, R. B. In *TAXOL® Science and Applications*; Suffness, M., Ed.; CRC Press: New York, 1995; pp. 259–274.
3. For a review, see: Ramos, A. C.; Peláez-Lamamié de Clairac, R.; Medarde, M. *Heterocycles* **1999**, *51*, 1443–1470.
4. Hitotsuyanagi, Y.; Yamagami, K.; Fujii, A.; Naka, Y.; Tahara, T. *J. Chem. Soc., Chem. Commun.* **1995**, 49–50; Hitotsuyanagi, Y.; Yamagami, K.; Fujii, A.; Naka, Y.; Ito, Y.; Tahara, T. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 1039–1042.
5. Yamagami, K.; Fujii, A.; Tahara, T.; Naka, Y.; Hitotsuyanagi, Y., unpublished results.
6. Tomioka, K.; Kubota, Y.; Koga, K. *Tetrahedron* **1993**, *49*, 1891–1900.
7. Ayres, D. C.; Pauwels, P. J. S. *J. Chem. Soc.* **1962**, 5025–5030.
8. All attempts to improve the selectivity of this reduction were unsuccessful due to ready migration and/or reductive elimination of the protective ethoxycarbonyl groups.
9. The stereochemistry of diols **9** and **10** was determined by chemical evidence. Diol **9** was subjected to hydrogenolysis, and successive dehydration under Mitsunobu conditions, which proceeds through an *anti* E2 mechanism, gave *Z*-olefin **20**. In the same manner, *E*-olefin was obtained from diol **10**.



Iimori, T.; Ohtsuka, Y.; Oishi, T. *Tetrahedron Lett.* **1991**, *32*, 1209–1212.

10. Lindgren, B. O.; Nilsson, T. *Acta Chem. Scand.* **1973**, *27*, 888–890; Kraus, G. A.; Taschner, M. J. *J. Org. Chem.* **1980**, *45*, 1175–1176.
11. Ninomiya, K.; Shioiri, T.; Yamada, S. *Tetrahedron* **1974**, *30*, 2151–2157.
12. Garegg, P. J.; Samuelsson, B. *J. Chem. Soc., Perkin Trans. 1* **1980**, 2866–2869.
13. Ley, S. V.; Norman, J.; Griffith, W. P.; Marsden, S. P. *Synthesis* **1994**, 639–666.

14. Compound **4**: Amorphous powder, $[\alpha]_D^{24}$ −168.8 (*c* 0.10, $CHCl_3$); ^1H NMR (500 MHz, C_5D_5N, ref: (C_5D_4HN)=7.21 ppm, *J*/Hz) δ 6.88 (s, 2H), 6.75 (s, 1H), 6.73 (s, 1H), 6.56 (d, 1H, *J*=1.5), 5.94 (d, 1H, *J*=1.2), 5.91 (d, 1H, *J*=1.2), 4.74 (d, 1H, *J*=5.0), 4.43 (dd, 1H, *J*=7.5, 6.5), 4.17 (m, 1H), 4.07 (dd, 1H, *J*=9.7, 7.5), 3.87 (s, 3H), 3.70 (s, 6H), 3.33 (dd, 1H, *J*=12.8, 5.0); ^{13}C NMR (125 MHz, C_5D_5N, ref: 135.5 ppm) δ 173.43 (s), 153.35 (s), 148.20 (s), 141.43 (s), 141.26 (s), 138.71 (s), 137.99 (s), 115.24 (s), 110.84 (d), 109.06 (d), 101.34 (t), 98.27 (d), 71.21 (t), 60.49 (q), 56.10 (q), 49.13 (d), 47.76 (d), 42.84 (d).
15. Although one of the referees urged us to add the results of the in vivo biological evaluation of analogue **4** to enhance the value of this manuscript, we decided to disclose the salient points of our study at this stage since the preparation of a sufficient amount of the analogue and such in vivo biological evaluation would involve an extended period of time, inevitably delaying the urgent publication of this synthetic work.