Tetrahedron Letters,Vol.30,No.22,pp 2953-2954,1989 0040-4039/89 $3.00 + .00
Printed in Great Britain Pergamon Press plc

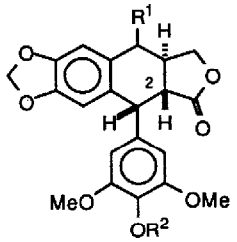
SYNTHESIS AND ANTITUMOR ACTIVITY OF PODOPHYLLOTOXIN AZA-ANALOGUES

Kiyoshi Tomioka,* Yoshihiro Kubota, and Kenji Koga*
Faculty of Pharmaceutical Sciences, University of Tokyo, Hongo, Bunkyo-ku, Tokyo 113, Japan

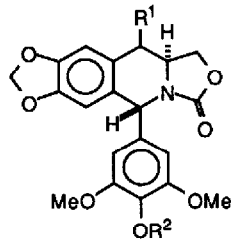
Summary: Podophyllotoxin aza-analogue 7 (3 R^1=H, R^2=Me, H) was designed and synthesized by a highly stereoselective condensation reaction of a cyclic urethane 5 with 3,4,5-trimethoxybenzaldehyde 6 and was found to show a promising in vitro and in vivo antitumor activity.

Podophyllotoxin **1** has a long and fascinating history as medicinals and this has recently culminated in the semi-synthetic analogues of clinically useful anticancer drugs **2** (teniposide and etoposide).[1,2] Since isomerization of cytotoxic **1** to inactive picropodophyllin is suggested to occur *via* epimerization at the C2 center under physiological conditions,[3] it is quite interesting to explore a new podophyllotoxin analogues which are incapable to lose configurational integrity at the C2 center. We report here design, synthesis, and antitumor activity of podophyllotoxin aza-analogues **3**.

Aza-analogues **3** have a sp^2 nitrogen at the corresponding C2 center of **1** and therefore epimerization at this center can be avoided.[4,5] From synthetic view point, **3**, both in racemic and optically pure forms, would be prepared in a quite short step starting from the known amino acid **4** *via* condensation of a cyclic urethane **5** and 3,4,5-trimethoxybenzaldehyde **6**.



1 R^1= α-OH R^2= Me
2 R^1= β-O-sugar R^2= H



3 R^1= H, OH
R^2= Me, H

A cyclic urethane **5** was prepared from a racemic **4** in 62% two-step yield. Condensation of **5** with 3,4,5-trimethoxybenzaldehyde **6** in the presence of H_2SO_4 (2 equiv) in CH_2Cl_2 at room temperature for 4 h provided a mixture of separable two diastereomers **7** (3 R^1=H, R^2=Me)(mp 185-186 °C) and **8** (mp 237.5-238.5 °C) in 93 and 3% yields, respectively. The structure was determined by observing NOE (in $CDCl_3$). An enhancement of 8% was observed between a proton of trimethoxyphenyl ring (δ 6.47) and a methine proton at the C3 center (δ 4.03) of the major product and none of enhancement was observed between the corresponding protons (δ 6.50 and 4.12) of the minor product, indicating **7** and **8** as major and minor products. It is quite important to note that *trans*-**7** was formed predominantly, in sharp contrast to the podorhizol cyclization[6a] and Pictet-Spengler reaction[6b] providing a *cis*-product.

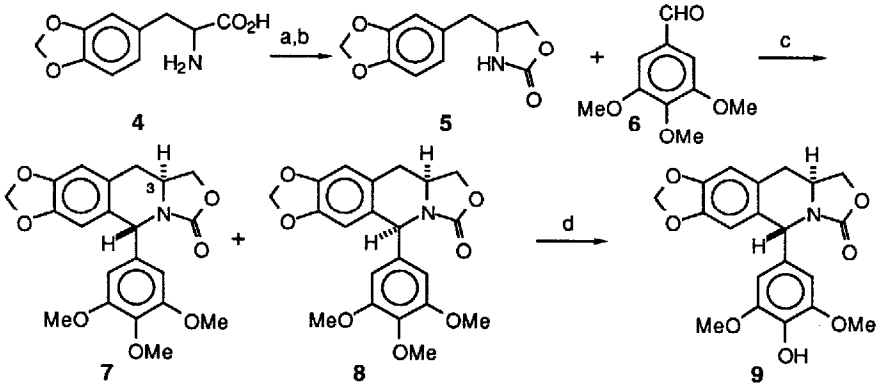
Treatment of **8** with acid (H_2SO_4/CH_2Cl_2, HBr/CH_2Cl_2, CF_3CO_2H/benzene, etc.) established a constant equilibrium to afford a mixture of **7** and **8** in a ratio of 28:1 (determined by HPLC analysis). In turn acid treatment of **7** also provided a mixture in the same ratio. These equilibrium between **7** and **8** is considered to occur *via* an intermediate **10** formed by C1-N bond cleavage. A trimethoxyphenyl ring of **8** is oriented pseudo-equatorial and sterically unfavourably interacted with C=O and C8-H

bonds on a plane, being epimerized to a pseudo-axial position in **7**. Predominant formation of **7** by equilibration rationalizes the highly stereoselective condensation reaction of **5** with **6**.

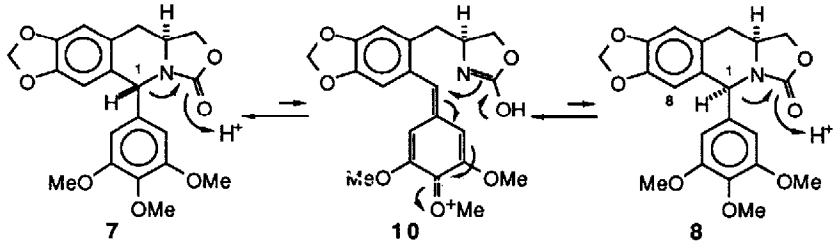
4'-Demethoxy derivative **9** (mp 210-212 °C) was prepared in 80% yield from **7** or **8** by treating with HBr in $Cl(CH_2)_2Cl$ at 0 °C for 14 h.

To our delightful **7**, **8**, and **9** exhibited promising growth inhibition of KB cell (ED_{50} <0.3 μg/ml) and *in vivo* activity against P-388 mouse (T/C 145 (**7**) and 170 (**8**)).

Further studies including synthesis and activity evaluation of optically active **3** (R^1=O-sugar, R^2=Me, H) will be reported in a forthcoming article.[7]



a) $LiAlH_4$/THF, reflux 2 h, 70%; b) $OC(OEt)_2$-NaOEt/EtOH, reflux 4 h, 89%; c) 3,4,5-trimethoxybenzaldehyde-H_2SO_4/CH_2Cl_2, rt 4 h, 93% for **7** and 3% for **8**; d) $HBr/Cl(CH_2)_2Cl$, 0 °C 14 h, 80%.



References and Notes

1. I. Jardin, in "Anticancer Agents Based on Natural Product Models," ed. by J. M. Cassady and J. D. Douros, 1980, Academic Press, p 319.
2. Synthesis of **1**: W. J. Gensler and C. D. Gatsonis, *J. Org. Chem.*, **31**, 4004 (1966); A. S. Kende, M. L. King, and D. P. Curran, *Ibid.*, **46**, 2826 (1981); D. Rajapaksa and R. Rodorigo, *J. Am. Chem. Soc.*, **103**, 6208 (1981); W. S. Murphy and S. Wattanasin, *J. Chem. Soc. Parkin I.*, **1982**, 271; J. Van der Eycken, P. De Clercq, and M. Vandewalle, *Tetrahedron*, **42**, 4297 (1986); D. M. Vyas, P. M. Skonezny, T. A. Jenks, T. W. Doyle, *Tetrahedron Lett.*, **27**, 3099 (1986); M. E. Jung and G. T. Lowen, *Ibid.*, **27**, 5319 (1986); T. Kaneko and H. Wong, *Ibid.*, **28**, 517 (1987); D. W. Jones and A. M. Thompson, *J. Chem. Soc., Chem. Commun.*, **1987**, 1797; D. I. MacDonald and T. Durst, *J. Org. Chem.*, **53**, 3663 (1988); R. C. Andrews, S. J. Teague, and A. I. Meyers, *J. Am. Chem. Soc.*, **110**, 7854 (1988).
3. H. Emmenegger, H. Stähelin, J. Rutschmann, J. Renz, and A. von Wartburg, *Arzneim.-Forsch*, **11**, 327, 459 (1961).
4. The same strategy was applied to steganine lignan. K. Tomioka, Y. Kubota, H. Kawasaki, and K. Koga, preceding paper.
5. Quite recently the similar approaches have been disclosed. H. L. Pearce, N. J. Bach, and T. L. Cramer, *Tetrahedron Lett.*, **30**, 907 (1989); M. Vandewalle, et al., *Japan Patent Kokai*, #283960 (1987).
6. a) K. Tomioka, T. Ishiguro, and K. Koga, *Chem. Pharm. Bull.*, **33**, 4333 (1985); b) M. Konda, T. Shioiri, and S. Yamada, *Ibid.*, **23**, 1025 (1975).
7. We are grateful to Drs. S. Tsukagoshi, T. Tsuruo, T. Tashiro, Cancer Institute, for activity evaluation.

(Received in Japan 15 March 1989)