Pergamon

Tetrahedron Letters 39 (1998) 7021–7024

TETRAHEDRON LETTERS

Model Studies for the Synthesis of the Marine Hepatotoxin Cylindrospermopsin. Preparation of a Bicyclic Guanidine with the Hydroxymethyluracil Side Chain.

Barry B. Snider* and Chaoyu Xie

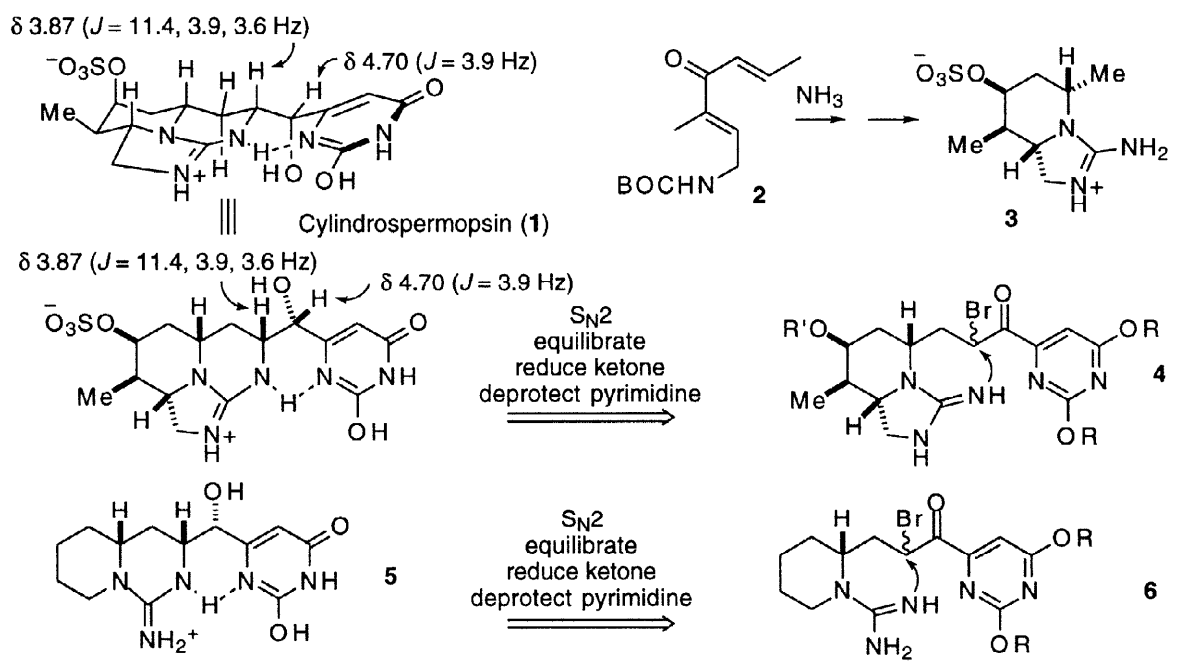
Department of Chemistry, Brandeis University, P. O. Box 9110,Waltham, MA 02254 (USA)

Received 15 June 1998; accepted 9 July 1998

Abstract: Ketone **13** was prepared by coupling acetylene **7b** with aldehyde **9** in a convergent six-step sequence. In the key step, hydrogenation of bromo ketone **14** afforded 81% of an 81:14:4.5:0.5 mixture of **15-18.** Hydrolysis of the major product **15** in concentrated hydrochloric acid at reflux afforded cylindrospermopsin model **5** in 95% yield. © 1998 Elsevier Science Ltd. All rights reserved.

Keywords: guanidines; pyrimidines, substitution

Cylindrospermopsin (**1**) was isolated from the cyanobacterium *Cylindrospermopsis raciborskii* by Moore and shown to be the causative agent of a 1979 outbreak of hepatoenteritis in Australia.[1] It has since been isolated from *Umezakia natans*[2a,b] and *Aphanizomenon ovalisporum.*[2c] Cylindrospermopsin is toxic to cultured rat hepatocytes and appears to function by inhibition of reduced glutathione synthesis.[3] The relative stereochemistry of the ring carbons of **1** was assigned based on the analysis of coupling constants. The stereochemistry of the side chain alcohol was tentatively assigned based on the coupling constants, NOEs, and the unusual behavior of the uracil carbons in the ^{13}C NMR spectrum, which suggested that the guanidinium is hydrogen bonded to the enol tautomer of the uracil as shown.



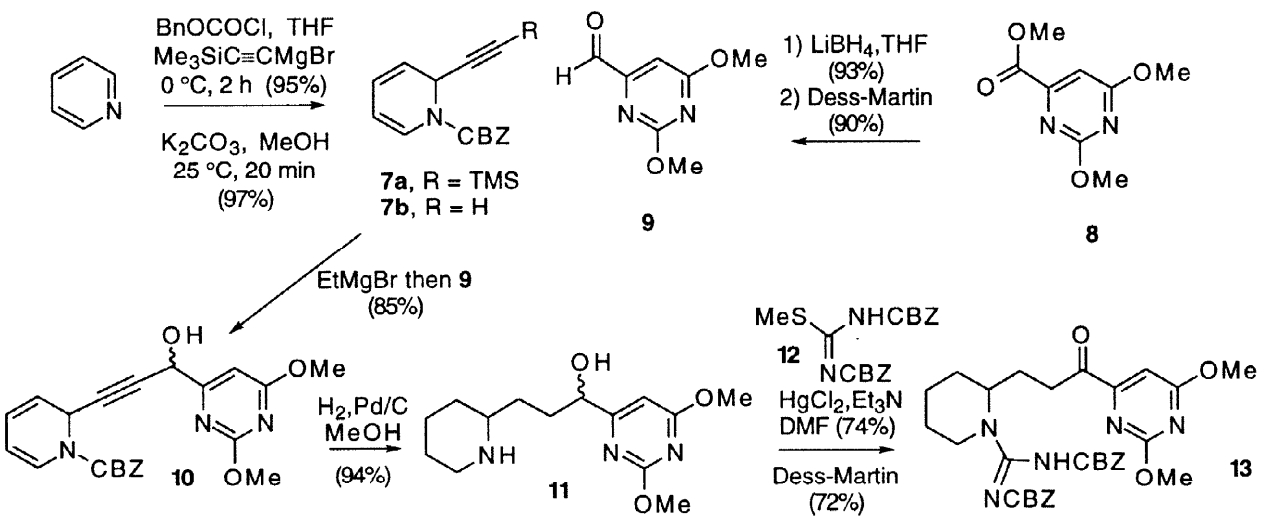
* E-Mail: snider@brandeis.edu; Fax: 781-736-2516

0040-4039/98/$ - see front matter © 1998 Elsevier Science Ltd. All rights reserved.
PII: S0040-4039(98)01470-1

The novel structure, with a guanidine embedded in a tricyclic system, and the profuse functionality makes the synthesis of cylindrospermopsin challenging.

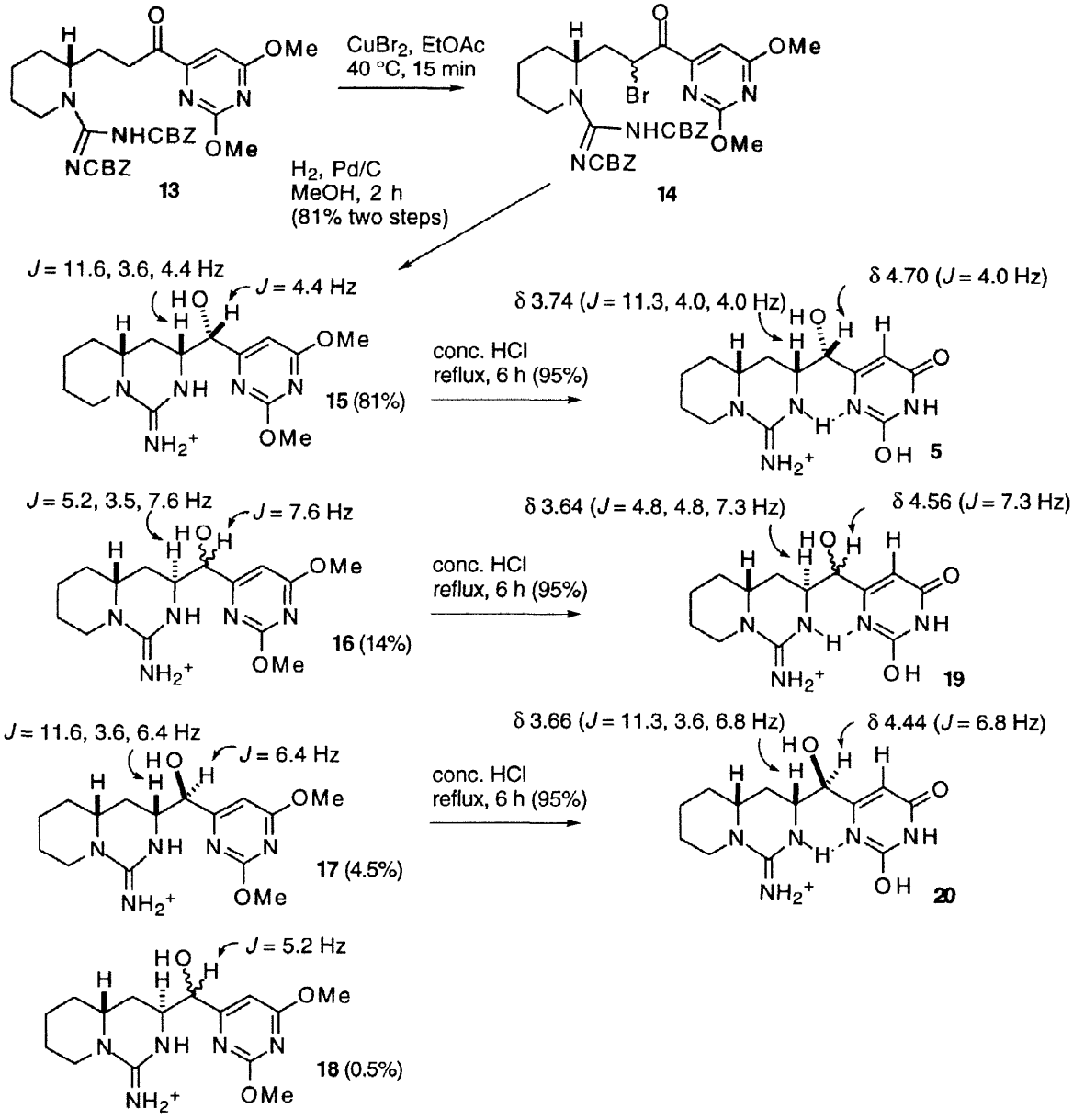
We have recently reported the synthesis of a bicyclic model for cylindrospermopsin in which the double Michael addition of NH_3 to dienone **2** formed a piperidone that was then elaborated to the bicyclic guanidinium sulfate **3**.[4,5] To complete the first synthesis of cylindrospermopsin (**1**), we needed to develop a procedure to form the third ring and the hydroxymethyluracil side chain. We hypothesized that bromo ketone **4**, which should be available analogously to **3**, could be elaborated to **1** by an S_N2 reaction to form the ring, equilibration to give the more stable equatorial side chain, reduction of the ketone to the alcohol, and liberation of the uracil. The formation of guanidine-containing rings by this procedure is novel, there are stereochemical concerns about the side chain equilibration and ketone reduction, and the optimal protecting group for the uracil needs to be experimentally determined. We therefore chose to investigate these steps in a model study with the simpler bromo ketone **6**, which should give cylindrospermopsin analog **5**.

A convergent approach to **6** was developed based on the coupling of acetylene **7b** with aldehyde **9**. Addition of $Me_3SiC{\equiv}CMgBr$ to pyridine and benzyl chloroformate by the procedure of Yamaguchi[6] afforded 95% of **7a** which was desilylated by treatment with potassium carbonate in MeOH at 25 °C to give 97% of terminal acetylene **7b**. Methyl 2,6-dimethoxy-4-pyrimidinecarboxylate (**8**) was prepared from orotic acid by the procedure of Gershon.[7] Although we were not sure that the methoxy groups could be cleaved, their ease of introduction, stability, and structural simplicity made them the optimal group for initial studies. Reduction of ester **8** with $LiBH_4$ in THF afforded 93% of the primary alcohol, which was oxidized to 90% of aldehyde **9**[8] with the Dess-Martin reagent. Treatment of **7b** with EtMgBr gave the acetylide, which was treated with aldehyde **9** to provide 85% of **10** as a mixture of diastereomers. Hydrogenation (1 atm) of **10** over 5% Pd/C reduced the dihydropyridine and acetylene, and cleaved the CBZ group affording 94% of **11** as a mixture of diastereomers. The guanidine was introduced in 74% yield by reaction of **11** with *N*,*N*'-bis(benzyloxycarbonyl)-*S*-methylisothiourea (**12**), Et_3N, and $HgCl_2$ in DMF.[9] Oxidation of the alcohol with the Dess-Martin reagent afforded 72% of ketone **13**.



Bromination of **13** with $CuBr_2$[10] in EtOAc for 15 min at 40 °C provided crude bromo ketone **14**, which was unstable and could not be purified. Fortunately, hydrogenolysis of

crude **14** (1 atm H_2, 5% Pd/C, MeOH, 2 h) liberated the free guanidine, which underwent an S_N2 reaction to form the second ring. Under these conditions, the ketone was also hydrogenated so that we obtained an 81:14:4.5:0.5 mixture of **15-18** in 81% yield from **13**. Careful flash chromatography on silica gel gave pure **15** and fractions containing **16** and **17** as the major product. The spectral data for **15** and **17** indicated that the hydroxymethyluracil side chain was equatorial. The ring methine hydrogen absorbs at δ 3.81 (ddd, J = 11.6, 3.6, 4.4 Hz) and δ 3.74 (ddd, J = 11.6, 3.6, 6.4 Hz) in **15** and **17**, respectively, with a large 11.6 Hz coupling constant between the axial methylene hydrogen and the axial methine hydrogen. The ring methine hydrogen of **16**, which absorbs at δ 3.65 (ddd, J = 5.2, 3.5, 7.6 Hz), must be equatorial since there is no large axial-axial coupling. The side chain methine hydrogen absorbs at δ 4.61 (J = 4.4 Hz) in **15**, and at δ 4.38 (J = 6.4 Hz) in **17**. Since this proton absorbs at δ 4.70 (J = 4.0 Hz) in **1**, the major product **15** probably has the same stereochemis-



try as cylindrospermopsin (**1**). However, the difference between the uracil of **1** and the dimethoxypyrimidine of **15** and **17** preclude definitive assignment at this point.

We therefore turned our attention to hydrolysis of the dimethoxypyrimidine to complete the synthesis of **5**. The dimethoxypyrimidine was resistant to hydrolysis, but was eventually cleaved in 95% yield by refluxing in concentrated hydrochloric acid for 6 h. We were delighted to find that no decomposition or isomerization occurred under these vigorous conditions. Apparently the protonated guanidine and uracil ring protect the alcohol from solvolysis, which would form a trication.

Hydrolysis of **15** gave **5**, with ^{1}H and ^{13}C NMR spectral data very similar to those of cylindrospermopsin (**1**), except for the expected differences in the left side of the molecule, while **20**, which was obtained from **17**, has very different spectral data.[11] For instance the absorption of the side chain methine hydrogen of **5** at δ 4.70 (J = 4.0 Hz) is identical to that of **1** and very different from that of **20** at δ 4.44 (J = 6.8 Hz). Therefore the major product of the S_N2 reaction and ketone hydrogenation sequence, **15**, has the same stereochemistry as the natural product. Although NOE studies on **5** and **20** support the literature assignment of the side chain alcohol stereochemistry, it must still be considered tentative.

We have developed a very short and efficient procedure for the formation of the tetrahydropyrimidine ring and hydroxymethyluracil side chain of cylindrospermopsin with good stereocontrol, which we are now applying to the synthesis of **1**.

Acknowledgement. We are grateful to the National Institutes of Health (GM-46470) for generous financial support.

References and Notes

1. (a) Ohtani, I.; Moore, R. E.; Runnegar, M. T. C. *J. Am. Chem Soc.* **1992**, *114*, 7941. (b) Moore, R. E.; Ohtani, I.; Moore, B. S.; De Koning, C. B.; Yoshida, W. Y.; Runnegar, M. T. C.; Carmichael, W. W. *Gazz. Chim. Ital.* **1993**, *123*, 329.
2. (a) Harada, K.; Ohtani, I.; Iwamoto, K.; Suzuki, M.; Watanabe, M. F.; Watanabe, M.; Terao, K. *Toxicon* **1994**, *32*, 73. (b) Terao, K.; Ohmori, S.; Igarashi, K.; Ohtani, I.; Watanabe, M. F.; Harada, K. I.; Ito, E.; Watanabe, M. *Toxicon* **1994**, *32*, 833. (c) Banker, R.; Carmeli, S.; Hadas, O.; Teltsch, B.; Porat, R.; Sukenik, A. *J. Phycol.* **1997**, *33*, 613.
3. (a) Runnegar, M. T.; Kong, S.-M.; Zhong, Y.-Z.; Ge, J.-L.; Lu, S. C. *Biochem. Biophys. Res. Comm.* **1994**, *201*, 235. (b) Runnegar, M. T.; Kong, S.-M.; Zhong, Y.-Z.; Lu, S. C. *Biochem. Pharm.* **1995**, *49*, 219.
4. Snider, B. B.; Harvey, T. C. *Tetrahedron Lett.* **1995**, *36*, 4587.
5. For other synthetic approaches see: (a) Heintzelman, G. R.; Parvez, M.; Weinreb, S. M. *Synlett* **1993**, 551. (b) Heintzelman, G. R.; Weinreb, S. M.; Parvez, M. *J. Org. Chem.* **1996**, *61*, 4594.
6. Yamaguchi, R.; Nakazono, Y.; Kawanisi, M. *Tetrahedron Lett.* **1983**, *24*, 1801.
7. Gershon, H. *J. Org. Chem.* **1962**, *27*, 3507.
8. Stogryn, E. L. *J. Heterocycl. Chem.* **1974**, *11*, 251.
9. (a) Chandrakumar, N. S; *Syn. Comm.* **1996**, *26*, 2613. (b) Tian, Z.; Edwards, P.; Roeske, R. W. *Int. J. Peptide Protein Res.* **1992**, *40*, 119. (c) Bergeron, R. J.; McManis, J. S. *J. Org. Chem.* **1987**, *52*, 1700. (d) Kim, K. S.; Qian, L. *Tetrahedron Lett.* **1993**, *34*, 7677.
10. Matsumoto, M.; Ishide, Y.; Watanabe N. *Heterocycles* **1985**, *23*, 165.
11. **5**: ^{1}H NMR (D_2O) 5.83 (s, 1), 4.70 (d, 1, J = 4.0), 3.80 (br d, 1, J = 12.8), 3.74 (ddd, 1, J = 11.3, 4.0, 4.0), 3.35 (dddd, 1, J = 11.0, 11.0, 4.6, 2.8), 2.94 (ddd, 1, J = 12.8, 12.8, 2.5), 2.06 (ddd, 1, J = 13.4, 4.6, 4.0), 1.92-1.75 (m, 3), 1.66 (ddd, 1, J = 13.4, 11.3, 11.0), 1.53-1.23 (m, 3); ^{13}C NMR (D_2O) 169.4, 158.0, 157.1, 155.3, 101.5, 71.7, 56.0, 53.7, 48.3, 34.6, 31.5, 26.7, 24.8; **19**: ^{1}H NMR (D_2O) 5.79 (s, 1), 4.56 (d, 1, J = 7.3), 3.64 (ddd, 1, J = 7.3, 4.8, 4.8), 3.35 (dddd, 1, J = 10.9, 10.9, 4.1, 4.0); **20**: ^{1}H NMR (D_2O) 5.84 (s, 1), 4.44 (d, 1, J = 6.8), 3.81 (br d, 1, J = 12.8), 3.66 (ddd, 1, J = 11.3, 6.8, 3.6), 3.37 (dddd, 1, J = 10.8, 10.8, 3.0, 3.0), 2.96 (ddd, 1, J = 12.8, 12.8, 2.6), 2.08 (ddd, 1, J = 13.6, 3.6, 3.0), 1.92-1.74 (m, 3), 1.68 (ddd, 1, J = 13.6, 11.3, 10.8), 1.58-1.22 (m, 3); ^{13}C NMR (D_2O) 169.5, 157.9, 157.0, 155.5, 101.9, 73.6, 56.1, 53.6, 48.3, 34.6, 34.0, 26.7, 24.8.