J|A|C|S
COMMUNICATIONS

Published on Web 01/05/2002

Total Synthesis of Ustiloxin D

Bin Cao, Haengsoon Park, and Madeleine M. Joullié*

Department of Chemistry, University of Pennsylvania, Philadelphia, Pennsylvania 19104-6323

Received October 10, 2001

In 1992, Iwasaki and co-workers reported the isolation of ustiloxin A (**1**) from the water extract of false smut balls on the panicles of the rice plant caused by the fungus *Ustilaginoidea virens*.[1] Later, four other congeners, ustiloxin B, C, D (**2**), and F, were isolated and characterized.[2] These natural products are 13-membered cyclic depsipeptides possessing a unique chiral tertiary alkyl−aryl ether linkage. Biological evaluation showed the ustiloxins to be potent antimitotic agents that strongly inhibit microtubule assembly by interfering with tubulin polymerization.[3] Due to the important role of microtubules in cell mitosis, compounds that bind tubulin and inhibit its assembly are potential anticancer drugs.[4] Ustiloxins have been shown to inhibit the growth of a variety of human tumor cell lines. To better understand the structure−activity relationships of tubulin binding, synthetic efforts by several groups have been directed toward a series of simplified ustiloxin model systems,[5] semisynthetic analogues,[6] the sulfinylnorvaline side chain of **1**,[7] and phomopsin A.[8] Herein, we describe the first total synthesis of ustiloxin D (**2**, Figure 1) in an effort to facilitate analogue preparation and provide structural variations for investigation of tubulin binding activity.

The main challenge in the total synthesis of **2** was the formation of the chiral tertiary alkyl−aryl ether linkage in the highly functionalized macrocycle. The key aspects of the synthetic strategy were the installation of the ether linkage at an early stage in order to circumvent future functional group incompatibility and the stereospecific generation of the β-hydroxytyrosine moiety in a single step. The retrosynthetic analysis of **2** is shown in Figure 1. Disconnection of the side chain as well as an amide bond within the macrocycle of **2** gave linear precursor **3**, which could be obtained via the regioreversed Sharpless asymmetric aminohydroxylation[9] of a cinnamate derivative. Further disconnections led to ether **4**. A number of methods have been reported for the synthesis of tertiary alkyl−aryl ethers, including nucleophilic substitution,[10] palladium-catalyzed cross-coupling,[11] and Mitsunobu reactions.[12] These approaches either failed to give the desired ether in ustiloxin D or were not suitable for the total synthesis. After extensive experimentation, we chose a nucleophilic aromatic substitution (S_NAr) approach to construct the ether linkage by reaction of nitrile-activated aryl fluoride **5** with tertiary alcohol **6**. Compound **6** could be prepared from D-serine.

The synthesis began with the preparation of amino alcohol **9** (Scheme 1). Alcohol **8** was synthesized from D-serine (**7**) in 5 steps.[13] Removal of the isopropylidene acetal gave a diol in which the liberated primary hydroxyl group was selectively protected as its MOM ether.[14] Subsequent removal of the Boc group afforded amino alcohol **9**. Treatment of **9** with **5** and KHMDS successfully introduced the ether linkage in **10** in 78% yield.[8a,15] The amino group was then protected as its Boc carbamate, and Raney nickel reduction converted **10** to aldehyde **11**.[16] Dakin oxidation converted **11** to its corresponding formate, and subsequent hydrolysis gave phenol **13**,[17] which was then protected as benzyl ether (**14**).

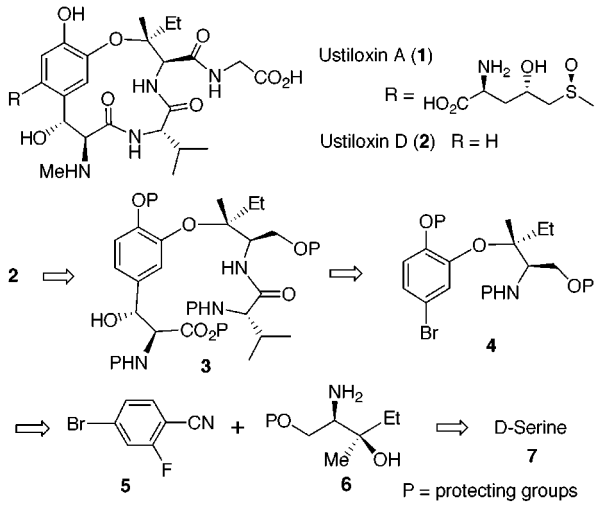


Figure 1. Retrosynthetic analysis of ustiloxin D.

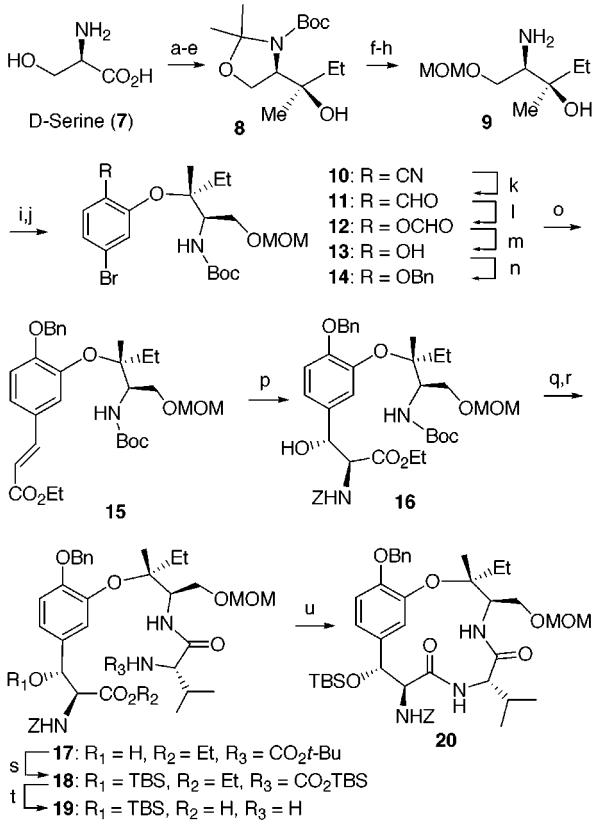
Treatment of **14** with ethyl acrylate under Heck conditions[18] afforded cinnamate **15** in 95% yield. The Sharpless regioreversed asymmetric aminohydroxylation protocol[9] was successfully applied to afford the desired (2*S*,3*R*)-β-hydroxy amino ester **16** in a single step (58% yield).[19] After removal of the Boc group, the amine was coupled with *N*-Boc-L-valine by using the DEPBT reagent.[20] Treatment of **17** with TBSOTf protected the secondary hydroxyl group as its silyl ether, as well as transformed the *tert*-butyl carbamate to the corresponding *tert*-butyldimethylsilyl carbamate.[21] Basic hydrolysis afforded amino acid **19**, which was then cyclized via macrolactamization to afford macrocycle **20** in 78% yield with EDCI−HOBt in a mixed solvent DMF−CH_2Cl_2 (1:5).[22]

With the macrocycle in hand, we were able to remove the MOM group (Scheme 2). To achieve selective methylation on the nitrogen of the tyrosine moiety, the benzyl carbamate was selectively removed by catalytic hydrogenolysis in the presence of the benzyl ether,[23] and amine **22** was subsequently protected as the 2-nitrobenzenesulfonamide **23**. Sequential treatment of the primary hydroxyl group in **23** with Dess−Martin reagent and then with $NaClO_2$ afforded a carboxylic acid, which was then coupled with glycine benzyl ester to give **24**. Selective deprotonation of the sulfonamide with MTBD[24] and subsequent methylation[25] before deprotection of the silyl ether afforded alcohol **25**. Removal of the 2-nitrobenzenesulfonyl group with β-mercaptoethanol gave a secondary amine. The final step involved hydrogenolysis with Pd black to remove both the benzyl ester and the benzyl ether to afford ustiloxin D (**2**) in 85% yield. The ^{1}H NMR, ^{13}C NMR, HRMS, and optical rotation of synthetic **2** were identical to those of the natural product.

In conclusion, the first total synthesis of **2** was accomplished from readily available D-serine and aryl fluoride starting materials. The synthesis provides a scaffold upon which we can construct other congeners and analogues to better understand the tubulin binding activity of the ustiloxin family of natural products.

10.1021/ja017277z CCC: $22.00

Scheme 1[a]

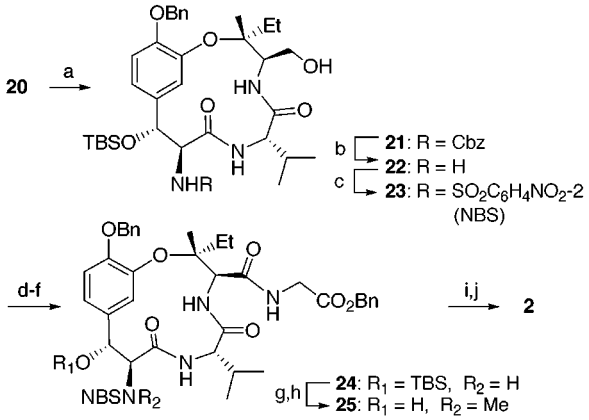


[a] Key: (a) Boc_2O, $NaHCO_3$, dioxane−H_2O; (b) MeNH(OMe)·HCl, EDCI·HCl, THF−H_2O, pH 4.5, 4 h, 71% for 2 steps; (c) $(MeO)_2CMe_2$, catalytic *p*-TsOH, 97%; (d) MeLi, THF, −78 to −65 °C, 72%; (e) EtMgBr, THF, −78 to −20 °C, ds = 9:1, 58%; (f) *p*-TsOH, MeOH; (g) MOMCl, *i*-Pr_2NEt, CH_2Cl_2, 0 °C to room temperature, 71% for 2 steps; (h) TFA, CH_2Cl_2, 0 °C, 78%; (i) 4-bromo-2-fluorobenzonitrile (**5**), 1.1 equiv of KHMDS, THF, 0 °C to room temperature, 78%; (j) Boc_2O, Et_3N, CH_2Cl_2, 81%; (k) Raney nickel, NaH_2PO_2, Pyr:H_2O:HOAc (2:1:1), 0−55 °C, 56%; (l) 30% H_2O_2, 4 mol % bis(2-nitrophenyl)diselenide, CH_2Cl_2, room temperature, 24 h; (m) KOH, MeOH−H_2O, 73% for 2 steps; (n) BnBr, K_2CO_3, *n*-Bu_4NI, DMF, 96%; (o) ethyl acrylate, $Pd(OAc)_2$, (*o*-tolyl)$_3P$, Et_3N, CH_3CN, reflux, 6 h, 95%; (p) NaOH, $BnOCONH_2$, *t*-BuOCl, $K_2[OsO_2(OH)_4]$, $(DHQD)_2AQN$, *n*-PrOH:H_2O (1:1), 20 °C, 1 h, 58%; (q) TFA, CH_2Cl_2, 0 °C; (r) *N*-Boc-L-valine·OH, DEPBT, *i*-Pr_2NEt, THF, 81% for 2 steps; (s) 6 equiv of TBSOTf, 8 equiv of 2,6-lutidine, CH_2Cl_2, room temperature; (t) LiOH, *t*-BuOH−H_2O, 83% for 2 steps; (u) 5 equiv of EDCI·HCl, 5 equiv of HOBt, DMF−CH_2Cl_2 (1:5), 0 °C to room temperature, 78%.

Acknowledgment. We thank the National Institutes of Health (CA-40081), the National Science Foundation (CHE-99-01449), and the University of Pennsylvania (Research Foundation) for financial support. We also thank Professor Shigeo Iwasaki and Dr. Yukiko Koiso for kindly providing samples of natural ustiloxins A and D.

Supporting Information Available: Experimental procedures and characterization data for the preparation of all new compounds and NMR spectra comparison of synthetic and natural **2** (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

Scheme 2[a]



[a] Key: (a) *B*-bromocatecholborane, CH_2Cl_2, −78 °C, 88%; (b) H_2, 5% Pd/C, 2,2′-dipyridyl, MeOH−EtOAc, 2 h; (c) 2-nitrobenzenesulfonyl chloride, 2,4,6-collidine, 85% for 2 steps; (d) Dess−Martin periodinane, CH_2Cl_2; (e) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH−H_2O; (f) Gly·OBn, DEPBT, *i*-Pr_2NEt, THF, 66% for 3 steps; (g) methyl 4-nitrobenzenesulfonate, MTBD, DMF, 77%; (h) *n*-Bu_4NF−HOAc (1:1.1), THF, 0−25 °C, 4 h, 96%; (i) $HSCH_2CH_2OH$, DBU, DMF, room temperature, 74%; (j) H_2, Pd black, EtOH−H_2O, 1.6 equiv of HCl, 2 h, 85%.

References

(1) Koiso, Y.; Natori, M.; Iwasaki, S.; Sato, S.; Sonoda, R.; Fujita, Y.; Yaegashi, H.; Sato, Z. *Tetrahedron Lett.* **1992**, *33*, 4157−4160.

(2) (a) Koiso, Y.; Li, Y.; Iwasaki, S.; Hanaoka, K.; Kobayashi, T.; Sonoda, R.; Fujita, Y.; Yaegashi, H.; Sato, Z. *J. Antibiot.* **1994**, *47*, 765−773. (b) Koiso, Y.; Morisaki, N.; Yamashita, Y.; Mitsui, Y.; Shirai, R.; Hashimoto, Y.; Iwasaki, S. *J. Antibiot.* **1998**, *51*, 418−422.

(3) (a) Ludueña, R. F.; Roach, M. C.; Prasad, V.; Banerjee, M.; Koiso, Y.; Li, Y.; Iwasaki, S. *Biochem. Pharmacol.* **1994**, *47*, 1593−1599. (b) Li, Y.; Koiso, Y.; Kobayashi, H.; Hashimoto, Y.; Iwasaki, S. *Biochem. Pharmacol.* **1995**, *49*, 1367−1372.

(4) (a) Hamel, E. *Med. Res. Rev.* **1996**, *16*, 207−231. (b) Iwasaki, S. *Med. Res. Rev.* **1993**, *13*, 183−198.

(5) For the construction of alkyl−aryl ether via the S_NAr approach, see: Laïb, T.; Zhu, J. *Synlett* **2000**, *9*, 1363−1365. Williamson alkylation approach, see: (a) Mutoh, R.; Shirai, R.; Koiso, Y.; Iwasaki, S. *Heterocycles* **1995**, *41*, 9−12. (b) Takahashi, M.; Shirai, R.; Koiso, Y.; Iwasaki, S. *Heterocycles* **1998**, *47*, 163−166.

(6) Morisaki, N.; Mitsui, Y.; Yamashita, Y.; Koiso, Y.; Shirai, R.; Hashimoto, Y.; Iwasaki, S. *J. Antibiot.* **1998**, *51*, 423−427.

(7) Hutton, C. A.; White, J. M. *Tetrahedron Lett.* **1997**, *38*, 1643−1646.

(8) Phomopsin A is an antimitotic agent whose structure is closely related to that of ustiloxins. See: (a) Woiwode, T. F.; Rose, C.; Wandless, T. J. *J. Org. Chem.* **1998**, *63*, 9594−9596. (b) Stohlmeyer, M. M.; Tanaka, H.; Wandless, T. J. *J. Am. Chem. Soc.* **1999**, *121*, 6100−6101. (c) Woiwode, T. F.; Wandless, T. J. *J. Org. Chem.* **1999**, *64*, 7670−7674.

(9) (a) Tao, B.; Schlingloff, G.; Sharpless, K. B. *Tetrahedron Lett.* **1998**, *39*, 2507−2510. (b) Park, H.; Cao, B.; Joullié, M. M. *J. Org. Chem.* **2001**, *66*, 7223−7226. We thank Professors Barry Sharpless and Valery Fokin for information on this asymmetric aminohydroxylation procedure.

(10) (a) Coutts, I. G. C.; Southcott, M. R. *J. Chem. Soc., Perkin Trans. 1* **1990**, 767−771. (b) Masada, H.; Oishi, Y. *Chem. Lett.* **1978**, 57−58.

(11) (a) Mann, G.; Hartwig, J. F. *J. Am. Chem. Soc.* **1996**, *118*, 13109−13110. (b) Palucki, M.; Wolfe, J. P.; Buchwald, S. L. *J. Am. Chem. Soc.* **1997**, *119*, 3395−3396. (c) Mann, G.; Hartwig, J. F. *J. Org. Chem.* **1997**, *62*, 5413−5418.

(12) (a) Subramanian, R. S.; Balasubramanian, K. K. *Synth. Commun.* **1989**, *19*, 1255−1259. (b) Bittner, S.; Assaf, Y. *Chem. Ind. (London)* **1975**, *6*, 281.

(13) Ageno, G.; Banfi, L.; Cascio, G.; Guanti, G.; Manghisi, E.; Riva, R.; Rocca, V. *Tetrahedron* **1995**, *51*, 8121−8134.

(14) Barton, D. H. R.; Géro, S. D.; Maycock, C. D. *J. Chem. Soc., Perkin Trans. 1* **1982**, *7*, 1541−1551.

(15) It is noteworthy that the reaction conditions do not tolerate an aryl fluoride bearing a benzylic proton *para* to the cyano group. When the amino group of **9** was protected as its *N*-carbamate, the undesired oxazolidinone was produced via intramolecular cyclization. With free amine **9** as the substrate, addition of excess base caused the formation of an undesired arylamine due to a faster reaction between in situ generated amide and aryl fluoride.

(16) Heffner, T. F.; Jiang, J.; Joullié, M. M. *J. Am. Chem. Soc.* **1992**, *114*, 10181−10189.

(17) Syper, L. *Synthesis* **1989**, *3*, 167−172.

(18) Cho, Y. J.; Rho, K. Y.; Keum, S. R.; Kim, S. H.; Yoon, C. M. *Synth. Commun.* **1999**, *29*, 2061−2068.

(19) Regioselectivity (5:1) and diastereoselectivity (91:9) were determined by HPLC on a Chiralpak AD column. The desired isomer can be separated from others by flash column chromatography.

(20) DEPBT: 3-(diethoxyphosphoryloxy)-1,2,3-benzotriazin-4(3*H*)-one. See: Li, H.; Jiang, X.; Ye, Y.-H.; Fan, C.; Romoff, T.; Goodman, M. *Org. Lett.* **1999**, *1*, 91−93.

(21) Sakaitani, M.; Ohfune, Y. *J. Org. Chem.* **1990**, *55*, 870−876.

(22) Boger, D. L.; Miyazaki, S.; Kim, S. H.; Wu, J. H.; Castle, S. L.; Loiseleur, O.; Jin, Q. *J. Am. Chem. Soc.* **1999**, *121*, 10004−10011. EDCI: 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide. HOBt: 1-hydroxybenzotriazole.

(23) Sajiki, H.; Hirota, K. *Tetrahedron* **1998**, *54*, 13981−13996.

(24) MTBD: 1,3,4,6,7,8-hexahydro-1-methyl-2*H*-pyrimido [1,2-*a*]-pyrimidine.

(25) Miller, S. C.; Scanlan, T. S. *J. Am. Chem. Soc.* **1997**, *119*, 2301−2302.

JA017277Z