Absolute Configuration and Total Synthesis of (+)-Epolactaene, a Neuritogenic Agent from *Penicillium* sp. BM 1689-P Active in Human Neuroblastoma Cells†

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Received January 20, 1998

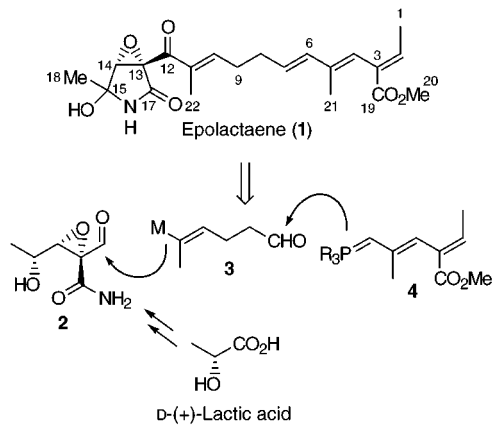
Neurotrophic factors including nerve growth factor (NGF)[1] stimulate the proliferation and differentiation of neuroblasts during their development and trigger adaptive plasticity responses in the mature nervous system. The ability of neurotrophic factors to control developmental neuronal survival and adult nervous system plasticity suggests the use of these molecules to treat neurodegeneration associated with human diseases.[2]

Epolactaene (**1**) was isolated as a diastereomeric mixture at the C-15 position (ca. 5:1 ratio) from the culture broth of *Penicillium* sp. BM 1689-P in 1995 by Osada et al.[3] The compound shows potent neurite outgrowth activity in the human neuroblastoma cell line SH-SY5Y.[4] Therefore, this natural product is noteworthy in the context of new drug development for various neurodegenerative diseases such as dementia.[5]

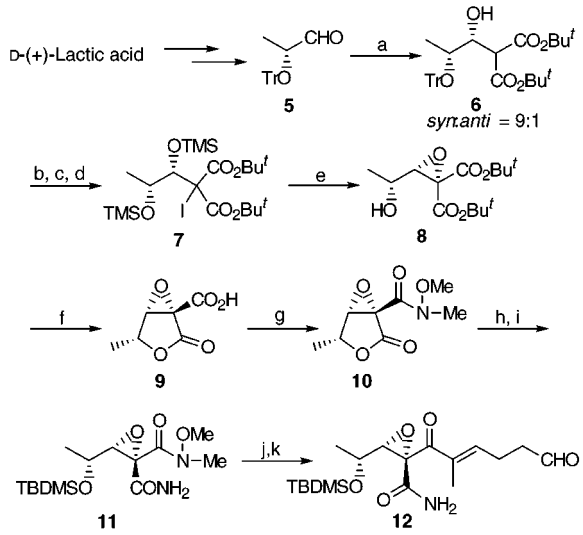
The gross structure of **1** was deduced by Osada using extensive NMR studies including $^1H-^1H$ COSY and HMBC. However, the initial structural assignment did not specify the absolute stereochemistry of the epoxy moiety, although it did establish the (*E*,*E*,*E*) geometry of the conjugated triene and the (*E*) configuration of the α,β-unsaturated ketone. We describe here the asymmetric total synthesis of (+)-epolactaene via a convergent approach that utilizes epoxyamide **2**, which is derived from D-(+)-lactic acid, C6 unit **3**, and Wittig reagent **4** (Scheme 1), followed by cyclization to form the lactam.

Readily available aldehyde (+)-**5**[6] ($[\alpha]^{22}_D$ +14.4° (*c* 0.50, $CHCl_3$)) derived from D-(+)-lactic acid was subjected to stereoselective aldol reaction with malonate anion to introduce a chiral center at the C-13 position of epolactaene (Scheme 2). The diastereoselective aldol reaction[7] was achieved by treating the aldehyde **5** with lithium malonate in the presence of $ZnCl_2$[8] at −78 °C to give the desired syn product **6** as 9:1 diastereomeric mixture in 53% yield[9] (85% yield based on 62% conversion of the aldehyde **5** by ^1H NMR analysis). After deprotection (80%) and silylation (94%) of **6**, the resulting bissilyl ether was purified by silica gel flash column chromatography to remove the minor anti isomer and then iodinated to obtain the α-iodo compound **7**. Subsequent desilylation with tetrabutylammonium fluoride (TBAF) gave the epoxide **8** in 52% yield (two steps). Treatment of this compound with formic acid provided epoxy lactone **9**, diastereoselectively generating a chiral center at the C-13 position. The lactone **9** was converted to the Weinreb amide[10] **10** by treating with (1*H*-benzotriazol-1-

Scheme 1



Scheme 2[a]



[a] Reagents and conditions: (a) (i) $ZnCl_2$, (ii) $LiCH(CO_2$-*t*-$Bu)_2$, THF, −78 °C (53%); (b) CF_3CO_2H, CH_2Cl_2, 0 °C (80%); (c) Me_3SiCl, imidazole, DMF, 0 °C (94%); (d) (i) LHMDS, (ii) I_2, −78 °C; (e) TBAF, THF, −45 to −15 °C (52% two steps); (f) HCO_2H, rt; (g) Me(MeO)NH·HCl, PyBOP, *i*-Pr_2NEt, CH_2Cl_2, 0 °C (69% from **8**); (h) NH_3, MeOH, rt; (i) TBDMSCl, imidazole, DMF, 0 °C to rt (93% from **10**); (j) (*Z*)-$Br(CH_3)C{=}CH(CH_2)_3OH$, *t*-BuLi, THF, −78 °C (83%); (k) Dess−Martin reagent, CH_2Cl_2, rt (81%).

† With regard to this investigation, a patent application was filed before Japanese Patent Office on Oct 30, 1997, as patent filing no. H9-297983.

(1) (a) Barde, Y, A.; *Neuron* **1989**, *2*, 1525−1535. (b) Ross, R. A.; Biedeler, J. L. *Cancer Res.* **1985**, *45*, 1628−1632.

(2) Hefti, F. *J. Neurobiol.* **1994**, *25*, 1418−1435.

(3) Kakeya, H.; Takahashi, I.; Okada, G.; Isono, K.; Osada, H. *J. Antibiot.* **1995**, *48*, 733−735.

(4) Kakeya, H.; Onozawa, C.; Sato, M.; Arai, K.; Osada, H. *J. Med. Chem.* **1997**, *40*, 391−394.

(5) Lactacystin has also been reported as the same neurite outgrowth activity. Isolation: (a) Omura, S.; Fujimoto, T.; Otoguro, K.; Matsuzaki, K.; Moriguchi, R.; Tanaka, H.; Sasaki, Y. *J. Antibiot.* **1991**, *44*, 113−116. (b) Omura, S.; Matsuzaki, K.; Fujimoto, T.; Kosuge, K.; Furuya, T.; Fujita, S.; Nakagawa, A. *J. Antibiot.* **1991**, *44*, 117−118. Synthesis: (c) Corey, E. J.; Reichard, G. A. *J. Am. Chem. Soc.* **1992**, *114*, 10677−10678. (d) Corey, E. J.; Reichard, G. A., Kania, R. *Tetrahedron Lett.* **1993**, *34*, 6977−6980. (e) Sunazuka, T.; Nagamitsu, T.; Matsuzaki, K.; Tanaka, H.; Omura, S.; Smith, A. B., III. *J. Am. Chem. Soc.* **1993**, *115*, 5302. (f) Uno, H.; Baldwin, J. E.; Russell, A. T. *J. Am. Chem. Soc.* **1994**, *116*, 2139−2140. (g) Chida, N.; Takeoka, J.; Tsutsumi, N.; Ogawa, S. *J. Chem. Soc., Chem. Commun.* **1995**, 793−794.

(6) Aldehyde (−)-**5** ($[\alpha]^{24}_D$ −14.4° (*c* 1.14, $CHCl_3$)) was synthesized by Mori et al.: Mori, K.; Kikuchi, H. *Liebigs. Ann. Chem.* **1989**, 963−967.

(7) For a review of stereoselective aldol reactions, see: (a) Franklin, A. S.; Paterson, I. *Contemp. Org. Synth.* **1994**, *1*, 317−338. (b) Heathcock, C. H. In *Comprehensive Organic Synthesis*; Trost, B. M., Fleming, I., Eds.; Pergamon: Oxford, 1991; Vol. 2, pp 181−238. (c) Heathcock, C. H. In *Asymmetric Synthesis*; Morrison, J. D., Eds.; Academic Press: New York, 1984; Vol. 3, pp 111−212. (d) Evans, D. A.; Nelson, J. V.; Taber, T. R. In *Topics in Stereochemistry*; Allinger, N. L., Eliel, E. L., Wilen, S. H., Eds.; John Wiley & Sons: New York, 1982; Vol. 13, pp 1−115.

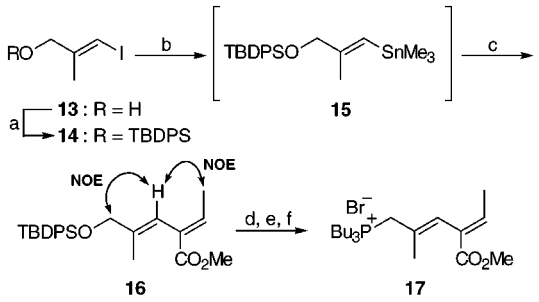
(8) No aldol product was obtained in the absence of $ZnCl_2$.

(9) The detailed study on this reaction will be described elsewhere.

(10) Nahm, S.; Weinreb, S. M. *Tetrahedron Lett.* **1981**, *22*, 3815−3818.

S0022-3263(98)00087-5 CCC: $15.00
Published on Web 03/11/1998

Scheme 3[a]

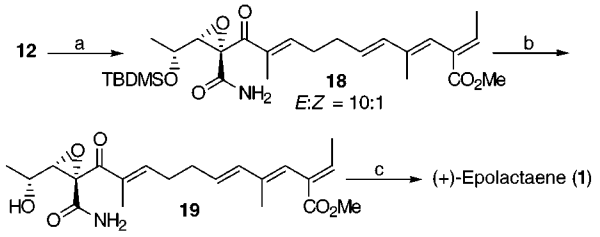


[a] Reagents and conditions: (a) *t*-BuPh$_2$SiCl, imidazole, DMF, rt (96%); (b) (i) *t*-BuLi, THF, − 78 °C, (ii) Me$_3$SnCl, THF, −78 °C; (c) (*Z*)-MeO$_2$C(Br)C=CHCH$_3$, Pd(Ph$_3$P)$_4$, toluene, reflux (47% from **14**); (d) TBAF, THF, 0 °C to rt (86%); (e) CBr$_4$, Ph$_3$P, THF, 0 °C (95%); (f) *n*-Bu$_3$P, CH$_3$CN, reflux (quantitative).

yloxy)tripyrrolidinophosphonium hexafluorophosphate[11] (PyBOP) and *N*,*O*-dimethylhydroxylamine in 69% overall yield from **8**. The absolute stereochemistry of C-13-C-14 epoxide and C-15 was assumed to be *cis* because the methine proton of the C-14 position of lactone **9** was observed as a doublet (J = 1.2 Hz).[12] The structure of **10** was firmly established by X-ray crystallographic analysis.[13] Exposure of **10** to ammonia in methanol followed by protection of alcohol with the *tert*-butyldimethylsilyl group provided the lactam precursor **11** in 93% yield from **10**. Coupling of vinyllithium, derived from (*Z*)-5-bromo-4-hexen-1-ol[14] with *tert*-butyllithium, with **11**, afforded the enone in 83% yield, which was then converted by Dess−Martin periodinane oxidation[15] to aldehyde **12** in 81% yield.

Wittig reagent **17** corresponding to the C-1 to C-6 unit was synthesized as shown in Scheme 3. (*E*)-3-Iodo-2-methyl-2-propen-1-ol (**13**)[16] was protected as the *tert*-butyldiphenylsilyl (TBDPS) ether to obtain **14** in 96% yield. Lithiation of **14** with *tert*-butyllithium followed by trapping with trimethyltin chloride afforded vinylstannane **15**, which was subjected to Stille coupling reaction[17] with methyl (*Z*)-2-bromo-2-butenoate[18] to furnish diene **16** in 47% yield. The stereochemistry of **16** was confirmed by NOE ^{1}H NMR studies. The silyl group of **16** was removed with TBAF (86%), and the resulting alcohol was transformed to the corresponding bromide with CBr$_4$ and triphenylphosphine (95%) followed by a treatment with tributylphosphine[19] to yield the desired Wittig reagent **17** in quantitative yield.

Scheme 4[a]



[a] Reagents and conditions: (a) **17**, 18-crown-6, *t*-BuOK, THF, −78 to −40 °C (69%); (b) 3HF·Et$_3$N, DMF, rt (96%); (c) Dess−Martin reagent, CH$_2$Cl$_2$, rt (74%).

Generation of the ylide from phosphonium salt **17** using potassium *tert*-butoxide in the presence of 18-crown-6 in THF followed by addition of aldehyde **12** resulted in the formation of the desired (*E*)-olefin **18** in high stereoselectivity (69% yield, *E*:*Z* = 10:1). The (*E*)-isomer was purified by silica gel flash column chromatography (Scheme 4), and its stereochemistry was confirmed by the large coupling constant (J_{6-7} = 15 Hz) observed in the ^{1}H NMR spectrum. Removal of the silyl group from **18** with triethylamine trihydrofluoride in *N*,*N*-dimethylformamide afforded alcohol **19** in 96% yield. Dess−Martin periodinane oxidation of **19** followed by spontaneous cyclization gave (+)-epolactaene (**1**) as an ca. 5:1 diastereomeric mixture at C-15, as with the natural product [[α]$^{22}_D$ +37° (*c* 0.2, MeOH) (lit.[3] [α]$^{22}_D$ +32° (*c* 0.1, MeOH))] in 74% yield. Synthetic (13*R*,14*R*)-(+)-epolactaene (**1**) exhibited physical and spectroscopic data (^{1}H NMR, ^{13}C NMR) identical with those of an authentic sample.

Acknowledgment. We thank Dr. Hiroyuki Osada, The Institute of Physical and Chemical Research (RIKEN), for copies of the NMR spectra of (+)-epolactaene. We also thank Dr. Tadashi Hata and Mr. Youji Furukawa, Analytical and Metabolic Research Laboratories, Sankyo Co. Ltd., for X-ray crystallographic analysis. This paper is dedicated to Prof. Isao Kuwajima (Tokyo Institute of Technology) on the occasion of his 60th birthday.

Supporting Information Available: Experimental details for the syntheses and spectroscopic data including a listing of crystallographic details for **10** (23 pages).

JO980087V

(11) Coste, J.; Le-Nguyen, D.; Castro, B. *Tetrahedron Lett.* **1990**, *31*, 205−208.
(12) Nishide, K.; Aramata, A.; Kamanaka, T.; Inoue, T.; Node, M. *Tetrahedron* **1994**, *50*, 8337.
(13) Crystal data: colorless prisms, orthorhombic $P2_12_12_1$, a = 9.577(2) Å, b = 15.361(1) Å, c = 6.609(2) Å, Z = 4, R = 0.041, R_w = 0.063. For full X-ray data, see the Supporting Information.
(14) This compound was synthesized from readily available (*E*)-3-bromo-2-buten-1-ol as follows: (a) MsCl, Et$_3$N, CH$_2$Cl$_2$, 0 °C; (b) NaH, CH$_2$(CO$_2$Et)$_2$, THF, 0 °C; (c) NaOH, EtOH, rt; (d) DMF, 100 °C; (e) K$_2$CO$_3$, MeI, DMF, rt; (f) LiAlH$_4$, THF, −78 °C (45% in overall yield). Corey, E. J.; Bock, M. G.; Kozikowski, A. P.; Rama Rao, A. V.; Floyd, D.; Lipshutz, B. *Tetrahedron Lett.* **1978**, *19*, 1051−1054.
(15) Dess, D. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4155−4156.
(16) Baker, R.; Castro, J. L. *J. Chem. Soc., Perkin. Trans. 1* **1990**, 47−65.
(17) (a) Stille, J. K. *Angew. Chem., Int. Ed. Engl.* **1986**, *25*, 508−524. (b) Farina, V.; Krishnamurthy, V.; Scott, W. J. *Org. React.* **1997**, *50*, 1−652.
(18) (a) Fevig, J. M.; Marquis, R. W., Jr.; Overman, L. E. *J. Am. Chem. Soc.* **1991**, *113*, 3, 5085−5086. (b) Hagiwara, H.; Abe, F.; Uda, H. *J. Chem. Soc., Perkin, Trans. 1* **1993**, 2651−2655.
(19) Tamura, R.; Saegusa, K.; Kakihana, M.; Oda, D. *J. Org. Chem.* **1988**, *53*, 2723−2728.