ORGANIC LETTERS

2002
Vol. 4, No. 10
1813−1815

Synthesis of Cryptophycin 52 Using the Shi Epoxidation

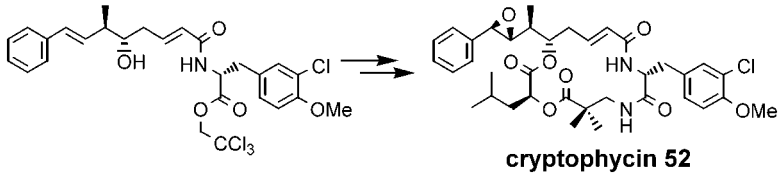
David W. Hoard,[†] Eric D. Moher,*[,†] Michael J. Martinelli,[†] and Bryan H. Norman[‡]

Chemical Process Research and Development and Discovery Chemistry Research Divisions, Lilly Research Laboratories, Eli Lilly and Company, Indianapolis, Indiana 46285

moher_eric_d@lilly.com

Received March 26, 2002

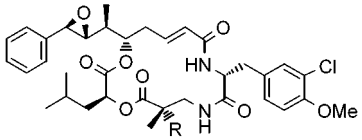
ABSTRACT



cryptophycin 52

A synthesis of cryptophycin 52 is reported using a Shi epoxidation strategy to install the epoxide moiety in a diastereoselective fashion. Several epoxidation results for cryptophycin substrates are disclosed followed by a discussion of the details relating to the preparation of cryptophycin 52 in two synthetic steps from one of the intermediate epoxides.

The cryptophycins,[1] exemplified by cryptophycin 1 (**1**), are cytotoxic macrocyclic depsipeptides isolated from blue-green algae (*Nostoc* sp. strains ATCC 537189[2] and GSV 224[3]). Cryptophycin 1 (**1**) has been shown by Moore and coworkers to be a potent tumor selective cytotoxin in vivo.[3,4] Noteworthy is the broad spectrum of antitumor activity exhibited by **1** across a variety of tumors implanted in mice. Particularly compelling is the observation that **1** significantly reduced the mean tumor burden from a Taxol-resistant mammary adenocarcinoma tumor implanted in mice.[3]



1: R = H
2: R = Me

As a result of these findings, we engaged in a collaboration with the University of Hawaii and Wayne State University aimed at discovering and developing cryptophycin analogues possessing refined biological properties. From this effort has emerged cryptophycin 52 (**2**),[5] which has undergone advanced clinical evaluation for the treatment of solid tumors.

Several research groups have adopted programs aimed at addressing the synthetic challenges associated with this structurally intriguing class of molecules. The first reports on the total synthesis of cryptophycins appeared in 1994 from the research groups of Kitigawa,[6] Moore,[7] and Tius.[7] These were soon followed by numerous approaches comprised of both formal and total syntheses.[8] A strategical theme common to the majority of syntheses of epoxide-containing crypto-

[†] Chemical Process Research and Development.
[‡] Discovery Chemistry Research.

(1) For recent reviews, see: (a) Shih, C.; Al-Awar, R. S.; Fray, A. H.; Martinelli, M. J.; Moher, E. D.; Norman, B. H.; Patel, V. F.; Shultz, R. M.; Toth, J. E.; Varie, D. L.; Corbett, T. H.; Moore, R. E. In *Anticancer Agents: Frontiers in Cancer Chemotherapy*; Ojima, I., Vite, G. D., Altman, K., Eds.; Washington, DC, 2001; Vol. 796, pp 171−189. (b) Eggen, M.-J.; Georg, G. I. *Med. Res. Rev.* **2002**, *22*, 85.

(2) Schwartz, R. E.; Hirsch, C. F.; Sesin, D. F.; Flor, J. E.; Chartrain, M.; Fromtling, R. E.; Harris, G. H.; Salvatore, M. J.; Liesch, J. M.; Yudin, K. *J. Ind. Microbiol.* **1990**, *5*, 113.

(3) Trimurtulu, G.; Ohtani, I.; Patterson, G. M. L.; Moore, R. E.; Corbett, T. H.; Valeriote, F. A.; Demchik, L. *J. Am. Chem. Soc.* **1994**, *116*, 4729.

(4) Golakoti, T.; Ogino, J.; Heltzel, C. E.; Husebo, T. L.; Jensen, C. M.; Larsen, L. K.; Patterson, G. M. L.; Moore, R. E.; Mooberry, S. L.; Corbett, T. H.; Valeriote, F. A. *J. Am. Chem. Soc.* **1995**, *117*, 12030.

(5) (a) Moore, R. E.; Tius, M. A.; Barrow, R. A.; Liang, J.; Corbett, T. H.; Valeriote, F. A.; Hemscheidt, T. K. PCT Int. Appl. WO 9640184 A1 19961219, 1996. (b) Wagner, M. M.; Paul, D. C.; Shih, C.; Jordan, M. A.; Wilson, L.; Williams, D. C. *Cancer Chemother. Pharmacol.* **1999**, *43*, 115 and references therein.

(6) Kobayashi, M.; Kurosu, M.; Wang, W.; Kitigawa, I. *Chem. Pharm. Bull. (Japan)* **1994**, *42*, 2394.

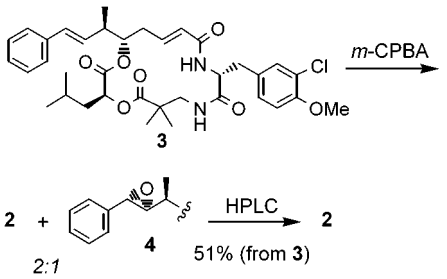
(7) Barrow, R. A.; Hemscheidt, T.; Liang, J.; Paik, S.; Moore, R. E.; Tius, M. A. *J. Am. Chem. Soc.* **1995**, *117*, 2479.

(8) For example, see: Eggen, M.; Nair, S. K.; Georg, G. I. *Org. Lett.* **2001**, *3*, 1813 and references therein.

10.1021/ol025933+ CCC: $22.00 © 2002 American Chemical Society
Published on Web 04/25/2002

phycins (e.g., **1**) has been the introduction of the epoxide pharmacophore in a single late-stage operation through the use of *m*-CPBA or dimethyl dioxirane. The reported epoxidations proceed with a diastereoselectivity of 2−3:1 necessitating a chromatographic separation of the desired (major) β-isomer. As part of our initial efforts to generate supplies of **2** and synthetic intermediates to fuel clinical development and SAR, respectively, we adopted the Moore−Tius synthesis of **2**.[5a,7] The final synthetic sequence[7,9] (Scheme 1)

Scheme 1



culminates in an epoxidation of styrene **3** employing *m*-CPBA to produce a 2:1 mixture of epoxides **2** and **4** from which **2** is isolated in 51% yield by reversed-phase HPLC. Herein, we report on an approach aimed at generating the epoxide of **2** in a more efficient manner to facilitate our support for continuing biological evaluation.

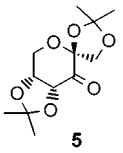
We began by screening general methods for the direct epoxidation of styrene **3** looking for enhanced diastereoselection relative to *m*-CPBA. Evident from the results depicted in Table 1, the only method giving any enhancement

Table 1. Reagent Screen for the Epoxidation of **3**

entry	reagent	time	temp	**2**:**4**[a]
1	*m*-CPBA	24 h	rt	1.9:1
2	$VO(acac)_2$- *t*-BuOOH	24 h	rt	1.9:1[b]
3	$Mo(CO)_6$- *t*-BuOOH	24 h	rt	1.9:1[b]
4[11]	Jacobsen (*S*,*S*)	23 h	rt	1:1.9[b]
5[11]	Jacobsen (*R*,*R*)	20 h	rt	1.9:1[b]
6	dimethyl dioxirane	4 h	0 °C	1.9:1
7	monoperoxyphthalic acid−Mg salt	24 h	rt	1.1:1
8[12]	$Ni(acac)_2$−RCHO−O_2	24 h	rt	c
9[13]	$MeReO_3$−H_2O_2	24 h	rt	1.7:1
10	CF_3CO_3H	1 h	0 °C	1.5:1
11	CH_3CO_3H	14 h	rt	1.7:1
12[14]	$TolSO_2$imidazole−H_2O_2	6 h	rt	c
13	*t*-BuOOH−$Ti(O\text{-}i\text{-}Pr)_4$	24 h	−10 °C	c
14[15]	$(EtO)_2POCl$−H_2O_2	8 h	rt	c
15	PhCN−H_2O_2	5 h	rt	c
16[10]	Shi method	4 h	rt	3.5:1[b]

[a] Ratio determined by HPLC; see ref 22. [b] Very low conversion to epoxides. [c] Styrene epoxidation products were not detected.

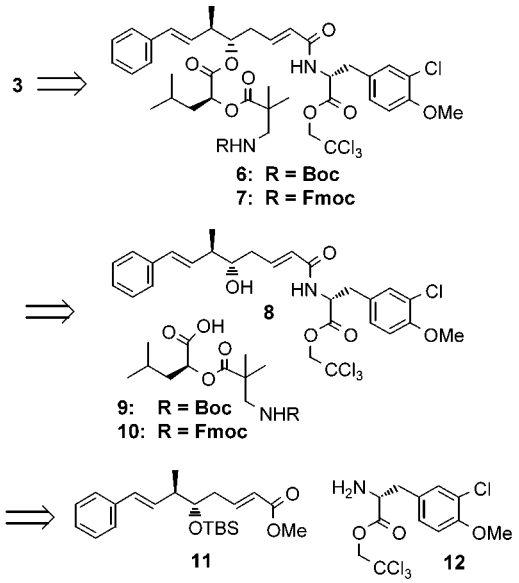
to the magnitude of diastereoselection was that of Shi (entry 16), which utilizes fructose-derived ketone **5** as a chiral dioxirane source.[10] While a promising hit, olefin **3** appeared to be a very poor substrate for the Shi epoxidation due to steric as well as solubility factors. A maximum conversion of only 17% was obtained under "forcing" conditions using 3 equiv of ketone **5** and 6 equiv of Oxone. Moreover, a mixed solvent system employing methylene chloride in addition to the prescribed acetonitrile or dimethoxymethane and water was necessary to help solubilize the substrate.



5

Having experienced very limited success in achieving a late-stage epoxidation with reasonable diastereocontrol, we turned our attention toward earlier intermediates in the synthesis, which may be more amenable to optimization of the Shi procedure. From careful inspection of the synthetic intermediates available from the Moore−Tius route, we identified styrenyl intermediate precursors to **3** that are the most compatible with an early epoxidation approach (Scheme 2). These substrates were subjected to an epoxidation screen

Scheme 2



using the Shi method as well as *m*-CPBA (Table 2).[16] The Shi epoxidation method offered the greatest level of dia-

(9) Fray, A. H. *Tetrahedron: Asymmetry* **1998**, *9*, 2777.
(10) Tu, Y.; Wang, Z.-X.; Shi, Y. *J. Am. Chem. Soc*. **1996**, *118*, 9806.
(11) Jacobsen Asymmetric Epoxidation (JAE): Jacobsen, E. N.; Zhang, W.; Muci, A. R.; Ecker, J. R.; Deng, L. *J. Am. Chem. Soc*. **1991**, *113*, 7063.
(12) Yamada, T.; Takai, T.; Rhode, O.; Mukaiyama, T. *Chem. Lett*. **1991**, 1.
(13) Adam, W.; Mitchell, C. *Angew. Chem., Int. Ed. Engl*. **1996**, *35*, 533.
(14) Kluge, R.; Schulz, M.; Liebsch, S. *Tetrahedron* **1996**, *52*, 2957.
(15) Kende, A. S.; Delair, P.; Blass, B. E. *Tetrahedron Lett*. **1994**, *35*, 8123.

Table 2. Shi[10] vs *m*-CPBA Epoxidation Results (β:α Epoxide Ratios) for Styryl Substrates

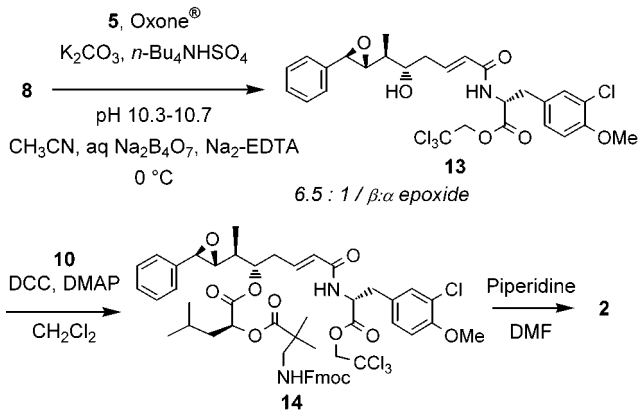
substrate	*m*-CPBA[a,b]	Shi[a,c]
11	1:1, >95% conversion	5:1, >95% conversion
8	1.5:1, >95% conversion[d]	5:1, >95% conversion
7	2:1, >95% conversion	9.5:1, 70−85% conversion

[a] Ratio determined by HPLC; see Supporting Information. [b] Methylene chloride, 1−2 equiv of 99% *m*-CPBA, room temperature. [c] Oxone (10 equiv), **5** (4 equiv), $NaHCO_3$ (aq), Na_2EDTA, Bu_4NOH, CH_3CN. [d] Buffered with aqueous sodium bicarbonate.

stereoselection observed to date for the epoxidation of the cryptophycin intermediates, while the *m*-CPBA cases gave levels of diastereoselection generally inferior to those exhibited by **3**. While modest, the selectivity generally observed across these substrates warranted further investigation into a synthetic route incorporating an early epoxidation strategy with the Shi epoxidation as a key step. All else being equal, late-stage intermediate **7** would be the preferred substrate for epoxidation since manipulations of the sensitive epoxide functionality would be minimized. However, the conversion to epoxide products was inferior to that of substrates **8** and **11** leaving behind significant levels of styrene starting material, which complicated the final purification of **2**. Indeed, obtaining high conversions for these substrates is a general challenge, the results of Table 2 reflecting the employment of 4 equiv of **5** and 10 equiv of Oxone for the epoxidations.

Further optimization was thus focused on styrene **8**. Gratifyingly, employing careful pH control[17] not only allowed for ketone **5** and Oxone levels to be reduced to 2 and 4 equiv, respectively, without compromising conversion, but a slight gain in diastereoselection to 6.5:1 β:α was realized (Scheme 3). With alcohol **13**[18] in hand, the stage was set for elaboration of the cryptophycin skeleton through esterification with an appropriately functionalized amino acid unit followed by deprotection and macrocyclization to furnish

Scheme 3



2. In the event, addition of crude **13** to a mixture of **10**,[18,19] DCC, and DMAP[20] furnished, after silica gel chromatography, fully protected amino ester **14**[18] in 71% overall yield[21] from styrene **8**. For the final stage of the synthesis, we envisioned cleavage of the Fmoc protecting group to reveal a free amine which, upon exposure to 2-hydroxypyridine as reported by Fray,[9] would smoothly afford macrocycle **2**. Toward that end, treatment of a DMF solution of **14** with 5 equiv of piperidine rapidly (<55 min) provided the expected seco-amine, which was not isolated but instead was observed to slowly convert to **2** in situ over 16 h giving cryptophycin 52 (**2**) in 79% isolated yield after chromatography.[21] Further purification of **2** to homogeneity, removing residual undesired α-epoxide **4**, could be readily accomplished through preparative reversed-phase HPLC in a manner analogous to that outlined in earlier reports.[22]

In conclusion, a synthesis of cryptophycin 52 (**2**) has been demonstrated based on the Shi epoxidation, which provides an efficient alternative method for installing the cryptophycin epoxide moiety. The route circumvents the commonly employed last-step epoxidation, which exhibits poor diastereoselectivity. An overall yield of 56% from styrene **8** was realized for this route as compared with 34% experienced for the Moore−Tius[5a,7] route using *m*-CPBA as the epoxidizing agent.

Acknowledgment. We thank Mr. David Robbins and Mr. Joseph Turpin for analytical support.

Supporting Information Available: Detailed experimental procedures and characterization for **13**, **14**, and **2**. This material is available free of charge via the Internet at http://pubs.acs.org.

OL025933+

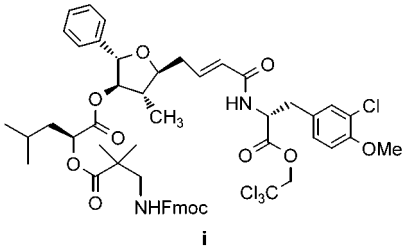
(16) These intermediates carry Fmoc nitrogen protection instead of the Boc protection used in the reported[5a,7] route. It was necessary to switch from Boc to Fmoc since the benzylic epoxide was not compatible with the acidic conditions necessary to remove the Boc group at a later point in the synthesis.

(17) Wang, Z.-X.; Yong, T.; Frohn, M.; Shi, Y. *J. Org. Chem.* **1997**, *62*, 2328.

(18) All isolated new compounds displayed ^1H NMR and mass spectral data consistent with their assigned structure.

(19) Fmoc-protected amine **10** was prepared from **9**[5a] in two steps: (a) HCl, EtOAc, 91%; (b) Fmoc−Cl, Na_2CO_3, dioxane, water, 93%.

(20) Premixing of all the components with DCC addition last, which is the typical protocol for DCC-mediated esterifications, led to the production of acylated tetrahydrofuran **i** as a byproduct (20%) contaminating **14**.



i

(21) Yield of β-epoxide, corrected for 10% α-epoxide present as a contaminant.

(22) (a) Patel, V. F.; Andis, S. L.; Kennedy, J. H.; Ray, J. E.; Schultz, R. M. *J. Med. Chem.* **1999**, *42*, 2588. (b) Also see ref 5a.