Organic LETTERS

Letter

pubs.acs.org/OrgLett

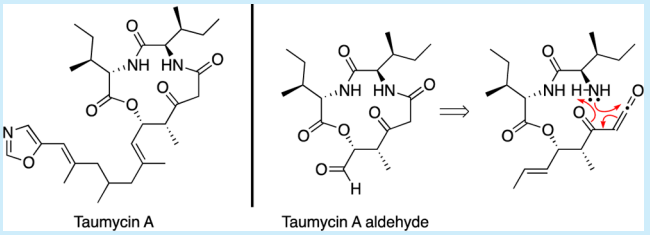
The Taumycin A Macrocycle: Asymmetric Total Synthesis and Revision of Relative Stereochemistry

Justine N. deGruyter and William A. Maio*

Department of Chemistry and Biochemistry, New Mexico State University, Las Cruces, New Mexico 88003, United States

Supporting Information

ABSTRACT: The first asymmetric total synthesis and revision of the relative configuration of the 12-membered taumycin A macrocycle is described. Key to the success of this work was a novel α-keto ketene macrocyclization that provided an efficient means by which to access two diastereomers of the desired macrolide without the need to employ additional coupling agents or unnecessary oxidation state adjustments.



The symbiotic association of marine microorganisms with their host sponges continues to be an extraordinary source of hybrid polyketide/nonribosomal secondary metabolites that often possess unique structures in conjunction with interesting biological activity.[1] Recently, taumycins A and B (**1**–**2**, Figure 1) were isolated by Kashman and co-workers, in extremely small amounts, from a Madagascar sponge of genus *Fascaplysinopsis*.[2a] Of particular note, taumycin A (**1**) was shown to inhibit UT-7 cell growth in the micromolar range, whereas taumycin B (**2**), which lacks the terminal oxazole moiety at C(12), is completely devoid of this activity.[2a] To date, extensive biological evaluation has not been performed on either metabolite, and the mechanism of action of **1** remains unknown.[2a]

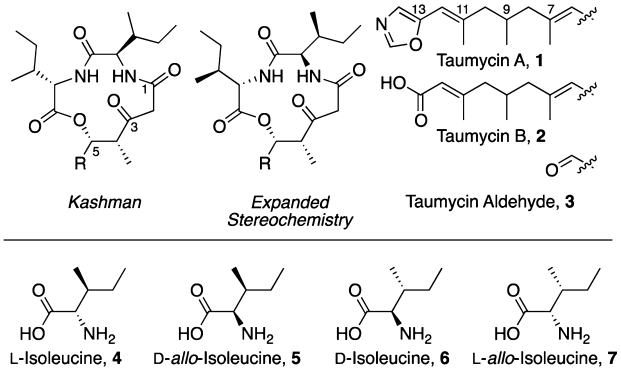


Figure 1. Taumycin family and stereochemistry comparison.

The relative configuration of the macrolide common to **1** and **2** was assigned using both detailed multidimensional NMR analyses and degradation studies (cf. **3**); however, due to its remote location, the configuration about the side chain C(9) methyl remains undefined. As determined by Kashman,[2a,b] the taumycin macrolide contains one L- and one D-isoleucine residue (Figure 1) which, in the absence of an X-ray quality crystal, made it difficult to establish absolute stereochemistry due to what was perceived to be potential ambiguity surrounding the correct sequence of amino acid subunits within the macrocycle.

We were attracted to taumycin A as a synthetic target not only because of its biological profile but also due to the reported presence of enantiomeric isoleucine residues. A careful examination of the isolation report, however, revealed that there was no clear discussion on how the data collected guided the authors to distinguish between D-isoleucine and D-*allo*-isoleucine (Figure 1).[3]

With this in mind, the relative stereochemistry as drawn by Kashman in the isolation report is somewhat unclear, as it does not adequately address the two stereocenters in each isoleucine residue (cf. Kashman vs. Expanded Stereochemistry, Figure 1). The presence of a D-amino acid is usually regarded as the empirical evidence that allows one to claim the source of a natural product to be bacterial derived, as it has been well established that certain bacteria are able to epimerize L-amino acids to their D-form.[4] In the case of isoleucine, however, α-epimerization would only affect the amino center, leaving the β-methyl center unchanged. Using this reasoning, L-isoleucine (**4**) can only be converted to its diastereomer, D-*allo*-isoleucine (**5**), and not its enantiomer, D-isoleucine (**6**). This is further supported by the fact that D-*allo*-isoleucine has been found previously in natural products,[3] while secondary metabolites which feature either D-isoleucine, or its α-epimer (**7**), are relatively uncommon in the literature.[5] We therefore believed that the taumycin macrolide was more likely to feature one L- and one D-*allo*-isoleucine (Figure 1).

At the onset of our synthetic campaign, we believed that the prudent approach might be to first target aldehyde **3**, a compound initially accessed by Kashman[2a] via reductive ozonolysis of **1**, as a means to address both the unknown

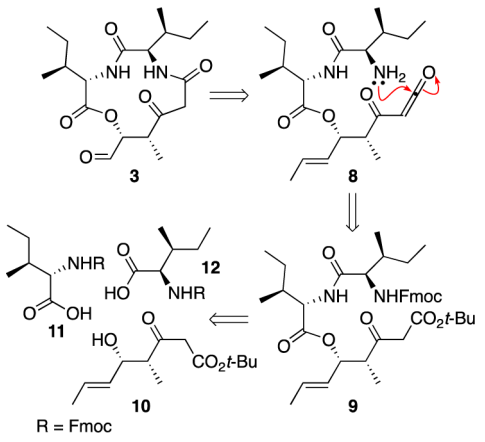
Received: August 29, 2014
Published: September 23, 2014

   dx.doi.org/10.1021/ol5025585 | *Org. Lett.* 2014, 16, 5196–5199

relative and absolute stereochemistry of the taumycin macrocycle. Once stereochemistry had been established, our plan was to then transition to the total syntheses of **1** and **2**, targeting both combinations of the undefined stereocenter at C(9).

From a retrosynthetic perspective (Scheme 1), we envisioned construction of the desired aldehyde in two sequential

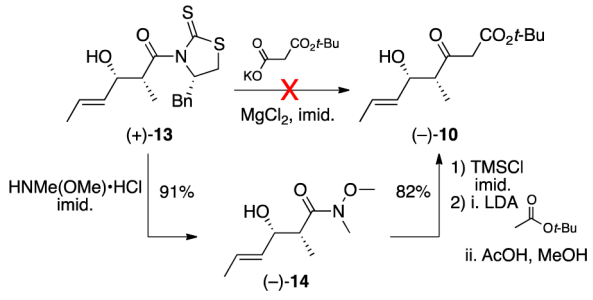
Scheme 1. Retrosynthetic Analysis



operations involving (1) macrocyclization followed by (2) alkene ozonolysis. Rather than rely on the use of unnecessary coupling reagents, we thought that a reasonable method would be cyclization exploiting an α-keto ketene (cf. **8**), which could be generated via the thermal rearrangement of β-keto *tert*-butylester **9**.[6] While similar approaches[7] have been documented for the synthesis of macrolides, to the best of our knowledge there has been no reported use of this strategy for depsipeptide cyclization.[8] Our rationale for pursuing this strategy came as a result of studying several reports on related depsipeptides, some noting difficulty in their macrocyclization steps; for example, the macrocyclization of a similar substrate was recently reported by Gosh and Xu[9] and required oxidation state adjustments to the 1,3-carbonyl moiety in order to cyclize. Ester **9** could in turn be synthesized via the stepwise union of amino acids **11** and **12** with alcohol **10**.

The synthesis of β-keto ester **10** commenced with the known non-Evans-*syn* aldol union between crotonaldehyde and 4-benzyl-3-propionyl-2-thiazolidine-2-thione, following the literature procedure of Crimmins,[10] to furnish (+)-**13** in good yield and high diastereoselectivity (Scheme 2). We chose to employ this titanium-mediated aldol method for several reasons: first, the ability to access both the Evans-*syn* and non-Evans-*syn* product from the same thiazolidinethione enantiomer by simply modulating the equivalents of base[10b,c] was extremely

Scheme 2. Synthesis of the C(1)−C(6) Fragment



appealing, since at the onset of our work it was still unclear which *syn* diastereomer of the southern hemisphere was needed for the natural product;[11] also of note, the ease with which the aldol product could be purified using silica gel chromatography, simply by collecting the bright yellow band as it elutes off the column, ensured that we could process decagram batches of pure material in an extremely time efficient manner; the most attractive feature, however, of the thiazolidinethione relative to other chiral auxiliaries (e.g., the Evans oxazolidinone) is its ability to be efficiently displaced by a variety of nucleophiles employing relatively mild reaction conditions.[12]

Initially, we thought that rapid access to β-keto ester **10** could be achieved through direct displacement of the thiazolidinethione using the potassium salt of mono-*tert*-butyl malonate and $MgCl_2$ (Scheme 2), similar to the elegant studies by Smith and co-workers employing the potassium salt of monoethyl malonate.[12a,b] Unfortunately, while we did observe auxiliary displacement, we could not prevent the *in situ* hydrolysis of the *tert*-butyl group under these conditions and any attempt led directly to the carboxylic acid derivative of **10**. Pleasingly, recourse to thiazolidinethione cleavage employing *N*,*O*-dimethylhydroxyl amine[12c] allowed efficient access to Weinreb amide (−)-**14** (Scheme 2). Alcohol protection followed by enolate displacement[13] and cleavage of the trimethylsilyl group upon workup afforded the desired C(1)−C(6) fragment [(−)-**10**] in good overall yield (58%). It should be noted that enolate displacement directly on nonprotected (−)-**14** gave good, but somewhat inferior, results compared to those in Scheme 2.

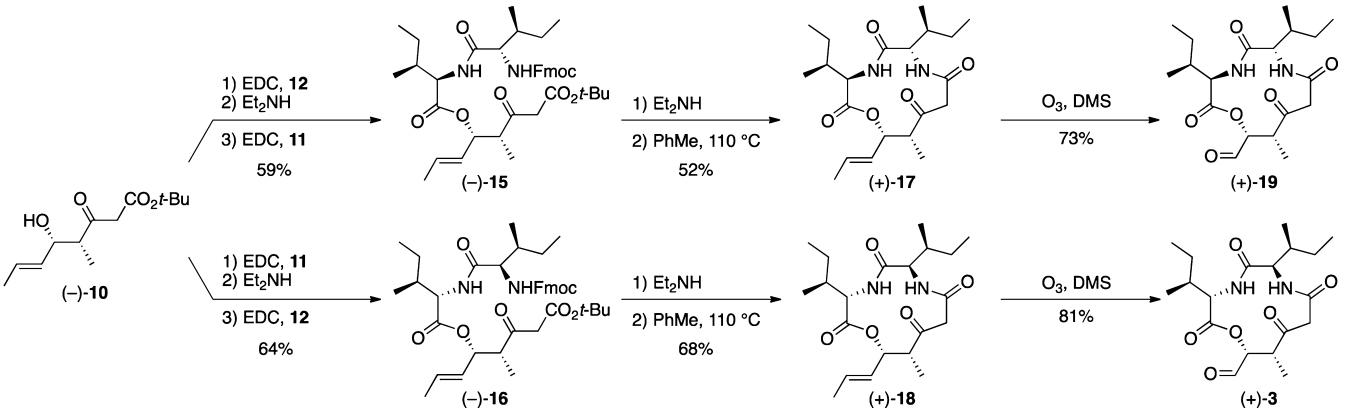
At this stage, we began our fragment union studies en route to synthetic taumycin A aldehyde. We knew that, in order to establish the correct relative configuration of **3**, it would be necessary to synthesize several macrocycles in which the order of amino acids would be varied (Scheme 3). We would also discover that the most efficient approach would not be the most convergent [i.e., uniting (−)-**10** with both dipeptide combinations].[14] Rather, we quickly determined that the iterative, three-step[15] addition of Fmoc protected amino acids[16] to sequentially install both combinations of the requisite dipeptide unit allowed for a more efficient route to access precyclization products (−)-**15** and (−)-**16** (Scheme 3). Notably, utilizing this strategy, the dipeptide unit can be installed in a single day, inclusive of intermediate purifications.[17,18]

With these compounds in hand, we next explored the viability of macrocyclization exploiting the thermal β-keto *tert*-butylester rearrangement. Based on the elegant studies of Witzeman and Nottingham[6a] regarding transacetoacetylations using *tert*-butylacetoacetate, as well as the seminal work by Boeckman[7a−c] and others[7d−o] on dioxinone rearrangements to form acylketene intermediates, we believed that thermal rearrangement in the presence of a primary amine [*e.g.*, derived after the Fmoc cleavage of either (−)-**15** or (−)-**16**], under high dilution conditions, should lead directly to the desired depsipeptide macrolides. To the best of our knowledge, there has been no reported use of the Witzeman method to effect macrocyclization, although the Boeckman method is a commonly employed tactic for ring closure.[7]

In the event, after Fmoc cleavage and gentle refluxing of the resultant amines for 2 h in toluene, macrocyclization to form amides (+)-**17** and (+)-**18** could be effected with excellent selectivity (mono vs poly) and in serviceable yield. Based on related observations by Hoye during his elegant synthesis of

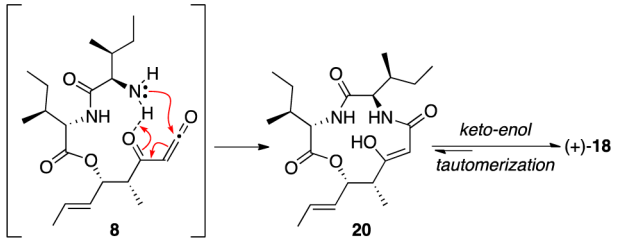
dx.doi.org/10.1021/ol5025585 | *Org. Lett.* 2014, 16, 5196−5199

Scheme 3. Synthesis of Aldehydes 19 and 3



callipeltoside,[19] we believe the transition from α-keto ketene to macrolide likely involves (1) carbonyl–N-H precoordination, followed by (2) the concerted movement of 6 π-electrons (cf. **8**, Scheme 4). In the final step, reductive ozonolysis of either (+)-**17** or (+)-**18** allowed access to aldehydes (+)-**19** and (+)-**3**, respectively.

Scheme 4. Proposed Transition State Precoordination



Pleasingly, there was a ^{1}H and ^{13}C NMR match between one of our synthetic derivatives and natural taumycin aldehyde [cf. (+)-**3**, Scheme 3]; this work has allowed us to confirm that the correct amino acid sequence is in the order L-isoleucine followed by D-*allo*-isoleucine. Our findings also contradict the initial claim that a D-isoleucine residue is present within the macrocycle and requires a revision to the relative configuration of the taumycin family to be made.[2a,b] Unfortunately, however, in the absence of a known optical rotation value for **3**, we are not able to definitively claim an absolute stereoassignment.

In conclusion, the relative stereochemistry of the taumycin macrolide has been confirmed via the first total synthesis of taumycin aldehyde (longest linear sequence is 10 steps from crotonaldehyde, 21% overall yield). Key to the success of this work was a novel macrocyclization tactic initiated by the thermal rearrangement of a β-keto *tert*-butylester. Full documentation of additional taumycin aldehyde diastereomers, including the D-isoleucine series, and studies directed toward the syntheses of taumycins A and B are ongoing and will be reported in due course.

■ ASSOCIATED CONTENT

Supporting Information

Detailed experimental procedures as well as scans of ^{1}H and ^{13}C NMR spectra are provided. This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Author

*E-mail: wmaio@nmsu.edu.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

This research was financially supported by the National Institutes of Health (SC2-GM099595-02). We are also grateful to the NIH Maximizing Access to Research Careers (MARC) program (NIH GM07667-36) for providing financial assistance to J.N.D.; to Professors Michael Crimmins (UNC), Junha Jeon (Arlington), Markus Kalesse (Leibniz Universität Hannover), Yoel Kashman (Tel Aviv, Emeritus), and Andrew G. Myers (Harvard) for providing helpful discussions; and to Professor John L. Wood (Baylor) for offering support as a mentor and for reviewing this manuscript. This manuscript is dedicated to Professor Moses K. Kaloustian with admiration and respect on the happy occasion of his 70th birthday in 2014.

■ REFERENCES

(1) (a) Walsh, C. T. *Science* **2004**, *303*, 1805–1810. (b) Hutchinson, C. R. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 3010–3012.

(2) (a) Bishara, A.; Rudi, A.; Aknin, M.; Neumann, D.; Ben-Califa, N.; Kashman, Y. *Org. Lett.* **2008**, *10*, 4307–4309. For related macrolides, see: (b) Bishara, A.; Rudi, A.; Goldberg, I.; Aknin, M.; Neumann, D.; Ben-Califa, N.; Kashman, Y. *Org. Lett.* **2009**, *11*, 3538–3541. (c) Ratnayake, R.; Fremlin, L. J.; Lacey, E.; Gill, J. H.; Capon, R. J. *J. Nat. Prod.* **2008**, *71*, 403–408. (d) Seo, C.; Yim, J. H.; Lee, H. K.; Park, S. M.; Sohn, J. H.; Oh, H. *Tetrahedron Lett.* **2008**, *49*, 29–31.

(3) For several recent examples, see: (a) Arrault, A.; Witczak-Legrand, A.; Gonzalez, P.; Bontemps-Subielos, N.; Banaigs, B. *Tetrahedron Lett.* **2002**, *43*, 4041–4044. (b) Kanoh, K.; Matsuo, Y.; Adachi, K.; Imagawa, H.; Nishizawa, M.; Shizuri, Y. *J. Antibiot.* **2005**, *58*, 289–292. (c) Matsuo, Y.; Kanoh, K.; Yamori, T.; Kasai, H.; Katsuta, A.; Adachi, K.; Shin-ya, K.; Shizuri, Y. *J. Antibiot.* **2007**, *60*, 251–255. (d) Portmann, C.; Blom, J. F.; Kaiser, M.; Brun, R.; Jüttner, F.; Gademann, K. *J. Nat. Prod.* **2008**, *71*, 1891–1896. (e) Ueoka, R.; Ise, Y.; Ohtsuka, S.; Okada, S.; Yamori, T.; Matsunaga, S. *J. Am. Chem. Soc.* **2010**, *132*, 17692–17694. (f) Tripathi, A.; Puddick, J.; Prinsep, M. R.; Rottmann, M.; Tan, L. T. *J. Nat. Prod.* **2010**, *73*, 1810–1814.

(4) (a) Bodansky, M.; Perlman, D. *Nature* **1968**, *218*, 291–292. (b) Cava, F.; Lam, H.; de Pedro, M. A.; Waldor, M. K. *Cell. Mol. Life Sci.* **2011**, *68*, 817–831.

(5) For a recent reported natural product with D-isoleucine, see: Takada, K.; Ninomiya, A.; Naruse, M.; Sun, Y.; Miyazaki, M.; Nogi, Y.; Okada, S.; Matsunaga, S. *J. Org. Chem.* **2013**, *78*, 6746–6750. For a L-

allo-isoleucine, see: Chen, Z.; Song, Y.; Chen, Y.; Huang, H.; Zhang, W.; Ju, J. *J. Nat. Prod.* **2012**, *75*, 1215−1219.

(6) (a) Witzeman, J. S.; Nottingham, W. D. *J. Org. Chem.* **1991**, *56*, 1713−1718. (b) Witzeman, J. S. *Tetrahedron Lett.* **1990**, *31*, 1401−1404. (c) Clemens, R. J.; Witzeman, J. S. *J. Am. Chem. Soc.* **1989**, *111*, 2186−2193.

(7) (a) Boeckman, R. K., Jr.; Perni, R. B. *J. Org. Chem.* **1986**, *51*, 5486−5489. (b) Boeckman, R. K., Jr.; Pruitt, J. R. *J. Am. Chem. Soc.* **1989**, *111*, 8286−8288. (c) Boeckman, R. K., Jr.; Weidner, C. H.; Perni, R. B.; Napier, J. J. *J. Am. Chem. Soc.* **1989**, *111*, 8036−8037. (d) Paquette, L. A.; Macdonald, D.; Anderson, L. G.; Wright, J. *J. Am. Chem. Soc.* **1989**, *111*, 8037−8039. (e) Chen, C.; Quinn, E. K.; Olmstead, M. M.; Kurth, M. J. *J. Org. Chem.* **1993**, *58*, 5011−5014. (f) Trost, B. M.; Gunzner, J. L. *J. Am. Chem. Soc.* **2001**, *123*, 9449−9450. (g) Trost, B. M.; Gunzner, J. L.; Dirat, O.; Rhee, Y. H. *J. Am. Chem. Soc.* **2002**, *124*, 10396−10415. (h) Cramer, N.; Laschat, S.; Baro, A.; Schwalbe, H.; Richter, C. *Angew. Chem., Int. Ed.* **2005**, *44*, 820−822. (i) Gebauer, J.; Blechert, S. *J. Org. Chem.* **2006**, *71*, 2021−2025. (j) Hart, A. C.; Phillips, A. J. *J. Am. Chem. Soc.* **2006**, *128*, 1094−1095. (k) Miyatake-Ondozabal, H.; Barrett, A. G. M. *Org. Lett.* **2010**, *12*, 5573−5575. (l) Simpson, T. J.; Soulas, F.; Willis, C. L. *Synlett* **2008**, 2196−2198. (m) Yoshinari, T.; Ohmori, K.; Schrems, M. G.; Pfaltz, A.; Suzuki, K. *Angew. Chem., Int. Ed.* **2010**, *49*, 881−885. (n) ElMarrouni, A.; Lebeuf, R.; Gebauer, J.; Heras, M.; Arseniyadis, S.; Cossy, J. *Org. Lett.* **2012**, *14*, 314−317. (o) Yadav, J. S.; Haldar, A.; Maity, T. *Eur. J. Org. Chem.* **2012**, 2062−2071.

(8) Davies, J. S. *J. Pept. Sci.* **2003**, *9*, 471−501.

(9) (a) Gosh, A. K.; Xu, C.-X. *Org. Lett.* **2009**, *11*, 1963−1966. Also see: (b) Trost, B. M.; Papillon, J. P. N.; Nussbaumer, T. *J. Am. Chem. Soc.* **2005**, *127*, 17921−17937.

(10) (a) Crimmins, M. T.; Chaudhary, K. *Org. Lett.* **2000**, *2*, 775−777. (b) Crimmins, M. T.; King, B. W.; Tabet, E. A.; Chaudhary, K. *J. Org. Chem.* **2001**, *66*, 894−902. (c) Crimmins, M. T.; She, J. *Synlett* **2004**, *8*, 1371−1374. For uses in total synthesis, see: (d) Crimmins, M. T.; DeBaillie, A. C. *J. Am. Chem. Soc.* **2006**, *128*, 4936−4937. (e) Crimmins, M. T.; Dechert, A.-M. R. *Org. Lett.* **2009**, *11*, 1635−1638.

(11) Through the course of our studies, it was necessary to synthesize all six stereocombinations in order to address the unknown configuration. In the interest of brevity, we have decided just to showcase the diastereomer series that led to the correct relative stereoisomer.

(12) (a) Smith, T. E.; Djang, M.; Velander, A. J.; Downey, C. W.; Carroll, K. A.; van Alphen, S. *Org. Lett.* **2004**, *6*, 2317−2320. (b) Tello-Aburto, R.; Olivo, H. F. *Org. Lett.* **2008**, *10*, 2191−2194. (c) Smith, A. B., III; Simov, V. *Org. Lett.* **2006**, *8*, 3315−3318.

(13) (a) Turner, J. A.; Jacks, W. S. *J. Org. Chem.* **1989**, *54*, 4229−4231. (b) Vosburg, D. A.; Vanderwal, C. D.; Sorensen, E. J. *J. Am. Chem. Soc.* **2002**, *124*, 4552−4553. (c) Vanderwal, C. D.; Vosburg, D. A.; Weiler, S.; Sorensen, E. J. *J. Am. Chem. Soc.* **2003**, *125*, 5393−5407.

(14) During initial studies, we observed an inefficient fragment union between an *N*-BOC-protected dipeptide with a fragment similar to (−)-**10**.

(15) For a good reference documenting the use of a carbodiimide to couple Fmoc protected amino acids and diethylamine to effect deprotection, see: Wen, S.; Packham, G.; Ganesan, A. *J. Org. Chem.* **2008**, *73*, 9353−9361.

(16) Fmoc-D-*allo*-ile-OH is commercially available from a variety of suppliers; however, we did not deem it cost-effective to purchase the large quantities required. Instead, we relied on a previously reported, two-step process to convert L-isoleucine into its D-*allo*-isomer, followed by nitrogen protection using FmocCl. For more information, see: (a) Noda, H.; Sakai, K.; Murakami, H. *Tetrahedron: Asymmetry.* **2002**, *13*, 2649−2652. (b) Yajima, T.; Horikawa, T.; Takeda, N.; Takemura, E.; Hattori, H.; Shimazaki, Y.; Shiraiwa, T. *Tetrahedron: Asymmetry* **2008**, *19*, 1285−1287. (c) Carpino, L. A.; Han, G. Y. *J. Org. Chem.* **1972**, *37*, 3404−3409.

(17) Several attempts were made to improve the coupling yields; however, after consulting the relevant literature, we found these yields to be comparable to previously reported studies that have employed Fmoc-protected amino acids. See: (a) Smietana, M.; Johnson, R. B.; Wang, Q. M.; Kool, E. T. *Chem.—Eur. J.* **2004**, *10*, 173−181. (b) Zlatopolskiy, B. D.; de Meijere, A. *Chem.—Eur. J.* **2004**, *10*, 4718−4727. (c) Fillion, D.; Deraët, M.; Holleran, B. J.; Escher, E. *J. Med. Chem.* **2006**, *49*, 2200−2209.

(18) Comparable results were obtained using DCC in place of EDC; however, the latter proved superior due to the ease of urea byproduct removal. The use of DIC, or coupling utilizing the α-azido acid derivatives of **11** or **12**, led to a significant amount of epimerization in our hands.

(19) (a) Hoye, T. R.; Danielson, M. E.; May, A. E.; Zhao, H. *J. Org. Chem.* **2010**, *75*, 7052−7060. Also see: (b) Freiermuth, B.; Wentrup, C. *J. Org. Chem.* **1991**, *56*, 2286−2289. (c) Birney, D. M.; Wagenseller, P. E. *J. Am. Chem. Soc.* **1994**, *116*, 6262−6270.