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Synthetic Studies toward the Mannopeptimycins: Synthesis of Orthogonally Protected β -Hydroxyenduracididines

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Abstract

The asymmetric synthesis of the nonproteinogenic amino acids (2*S*,3*S*,4'*S*)- β -hydroxyenduracididine 3 and (2*R*,3*S*,4'*S*)- β -hydroxyenduracididine 4 in orthogonally protected form in 15 total steps from Garner's aldehyde is reported. The former and *N*-glycosylated form of the latter are found in the glycopeptide antibiotic mannopeptimycin.

The rapidly escalating fight against bacterial resistance to currently available therapies has created an urgent need for new antibiotic agents with novel modes of action. For example, *Staphylococcus aureus*, which has a long history of acquiring resistance to antibiotics, continues to pose problems for existing chemotherapeutic agents. In fact, methicillin-resistant *S. aureus* (MRSA) infections have been a significant health problem since the 1960s.¹ A recent study estimated that there were over 94,000 cases of invasive MRSA infection in the United States in 2005.² While vancomycin has been an effective drug for treating these infections, recent identification of MRSA isolates possessing an intermediate level of resistance to vancomycin has underscored the need for novel antibiotics that act via mechanisms that are orthogonal to currently used therapies.¹

The mannopeptimycins are a group of cyclic glycopeptides originally isolated in the 1950s as an antibiotic complex produced by the LL-AC98 strain of *Streptomyces hygroscopicus*.³ They represent a novel class of lipoglycopeptide antibiotics that are assembled via a non-ribosomal peptide synthetase (NRPS) pathway and have shown promising antibacterial activity against multi-drug resistant pathogens.⁴ Only in 2002 was a report of the complete structural and stereochemical assignment of the five members of this complex disclosed. The structures of the mannopeptimycins and their biological activities are summarized in Table 1.

Mannopeptimycins β - ϵ are active against a wide range of Gram-positive bacteria, including methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Enterococcus*

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Supporting Information Available Experimental procedures and tabulated spectroscopic data for all new compounds. This material is available free of charge via the Internet at <http://pubs.acs.org>.

faecium.^{5,6} A semisynthetic analog of the mannopeptimycins, AC98-6446 has shown significantly greater potency than the natural mannopeptimycins against MRSA, VRE, penicillin-resistant *Streptococcus pneumoniae*, and glycopeptide intermediate *Staphylococcus aureus*.^{6,7,8} Mechanism of action studies have shown that mannopeptimycin interferes with the late stages of bacterial cell wall synthesis, most likely the transglycosylation reaction, possibly through binding to lipid II.⁹

The peptide core of the mannopeptimycins is composed of six amino acid residues of alternating D- and L-amino acids, three of which are nonproteinogenic. Two of these non-proteinogenic amino acids, the β -hydroxyenduracididines, have not been found in any other natural product although the biosynthetic pathway used for their construction is currently being studied.^{4,10} In addition, two of these non-proteinogenic amino acids (D-tyrosine and β -D-hydroxyenduracididine) are glycosylated with mannose residues; the former is glycosylated with an α -(1,4)-linked-bis-manno-pyranosyl pyranoside^{11,12} and the latter is *N*-glycosylated with an α -linked mannose residue.

An important element of any effort directed toward the total synthesis of mannopeptimycin is development of an efficient asymmetric synthesis of the novel β -hydroxyenduracididine residues **3** and **4** with appropriate orthogonal protecting groups. Each of these residues is densely functionalized—every carbon atom in the amino acid backbone is heteroatom-substituted—and features three contiguous stereocenters. Synthetic routes to either of these unusual amino acids have yet to be reported. Our interest in studying the influence of structure on the biological activity of the mannopeptimycins required an efficient and robust synthetic route to each of the β -hydroxyenduracididines. Herein we detail our synthetic route to these mannopeptimycin building blocks.

Given that the hydroxyenduracididines differ only in the stereochemical configuration at C-2, it was envisaged that they could be accessed from a common advanced intermediate. Our strategy, which is depicted in Scheme 1, involved the preparation of the α -nosylate ester **2**, which was subsequently manipulated to provide each of the hydroxyenduracididine diastereomers. Nosylate **2** could be accessed via Sharpless asymmetric dihydroxylation¹³ of an appropriately derivatized enoate precursor (e.g., **11**) followed by selective activation of the α -hydroxyl group via nosylation.¹⁴ The (2*S*, 3*S*, 4'*S*)-diastereomer **3** (L-absolute configuration) could be accessed via direct azide displacement of the nosylate group in **2**. Conversion of nosylate **2** to an epoxide intermediate followed by nucleophilic ring-opening by azide would provide access to the (2*R*, 3*S*, 4'*S*)-diastereomer **4** (D-absolute configuration).^{14a}

The synthesis of nosylate **2** commenced with olefin **6**,¹⁵ which was prepared by Wittig olefination of (*S*)-Garner's aldehyde. As detailed in Scheme 2, acidic cleavage of the *N,O*-acetonide and *N*-Boc protecting groups (6*N* HCl, 1h) was followed by guanidinylation with *N,N'*-bis-(benzyloxycarbonyl)-*S*-methylisothiourea **7** (HgCl₂, Et₃N, DMF, 4 d) to afford alcohol **8** in 45% overall yield from **6**. Cyclization of **8** under Mitsunobu conditions (DIAD, Ph₃P, THF, -15 °C) provided alkene **9** that contained the 2-iminoimidazolidine group in partially protected form.

At this juncture, orthogonal protection of the remaining nitrogen atom of the 2-iminoimidazolidine was required in order to insure that it could be selectively unmasked for subsequent glycosylation. Since the remainder of the synthetic route would leverage the Sharpless asymmetric dihydroxylation reaction and subsequent electrophilic activation/displacement reactions, the choice of protection for this nitrogen must satisfy several requirements. First, it must not contain electrophilic functionality capable of reacting with nearby hydroxyl groups that would be introduced with Sharpless asymmetric dihydroxylation. Second, it must not contain nucleophilic functionality that would be capable of displacing either

of the activated hydroxyl groups. Third, it must be selectively cleaved under mild conditions that will not also cleave the neighboring Cbz-protecting groups. These restrictions rule out a number of commonly used protecting groups (e.g., carbamates, amides, benzyl, etc.).

Given these limitations, we chose to protect the remaining nitrogen atom of **9** with the (triisopropylsilyloxy)methyl (Tom) group.¹⁶ Thus, olefin **9** was treated with Tom-Cl (KHMDs, toluene, 1 h) to provide the Tom-protected olefin **10** in modest yield. Olefin **10** was subjected to a dihydroxylation/oxidative cleavage sequence (OsO₄, NMO, THF/H₂O/*t*-BuOH then NaIO₄, CH₂Cl₂/H₂O) and the crude aldehyde was treated with methyl (triphenylphosphoranylidene)acetate at low concentration in THF to give enoate **11** in 71% overall yield and a 17:1 product ratio favoring the desired (*E*)-(*a*) Fleming, P. R.; Sharpless, K. B. *J. Org. Chem.* **1991**, *56*, 2869–2875. (AD-mix *α*, *t*-BuOH/H₂O) proceeded cleanly to provide a single diol diastereomer **12**. Monosulfonylation of diol **12** (Nos-Cl, Et₃N, CH₂Cl₂, –40 to 0 °C) proceeded smoothly to provide nosylate **2** in 71% yield.

The nosylate intermediate **2** was then transformed into orthogonally-protected (2*S*,3*S*,4'*S*)-diastereomer **3** (β-L-hydroxyenduracididine) in two steps (Scheme 3). Nucleophilic displacement of the nosylate in **2** with sodium azide (NaN₃, DMF, 50 °C, 78%) provided an intermediate azido alcohol which was subjected to a one-pot Staudinger reduction (Ph₃P, THF/H₂O, 4 h) and Boc protection (Boc₂O, NaHCO₃, 12 h) to provide building block **3** in 62% overall yield from the intermediate azido alcohol.

Whereas **3** was prepared via inversion of the stereochemical configuration at C-2, its epimer **4** was obtained by double inversion at that center, as outlined in Scheme 3. Conversion of **2** to epoxide **13** (KHMDs, THF/tol, –78 °C, 59%) followed by nucleophilic ring opening in the presence of sodium azide and pyridinium *p*-toluenesulfonate (DMF, 44%) afforded an intermediate azido alcohol which was converted to the orthogonally-protected (2*R*,3*S*,4'*S*)-diastereomer **4** (β-D-hydroxyenduracididine) via the aforementioned Staudinger reduction/Boc-protection procedure.

In conclusion, we have completed the first chemical synthesis of the (2*S*,3*S*,4'*S*)- and (2*R*,3*S*,4'*S*)-hydroxyenduracididines in orthogonally protected form. Access to each of these non-proteinogenic amino acids will assist efforts directed at total synthesis of members of the mannopeptimycin class of lipoglycopeptide antibiotics and aid semisynthetic efforts directed toward identification of mannopeptimycin derivatives with improved biological activity profiles. Furthermore, these novel amino acids will support efforts directed toward elucidating the mode of action and binding site(s) utilized by the mannopeptimycins. Our results from these studies will be presented in due course.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

1. Liñares J. *J Clin Microbiol Infect* 2001;7:8–15.
2. Kleven RM, Morrison MA, Nadle J, Petit S, Gershman K, Ray S, Harrison LH, Lynfield R, Dumyati G, Townes JM, Craig AS, Zell ER, Fosheim GE, McDougal LK, Carey RB, Fridkin SK. *JAMA* 2007;298:1763–1771. [PubMed: 17940231]

3. He H, Williamson RT, Shen B, Graziani EI, Yang HY, Sakya SM, Petersen PJ, Carter GT. *J Am Chem Soc* 2002;124:9729–9736. [PubMed: 12175230]
4. Magarvey NA, Haltli B, Greenstein M, Hucul JA. *Antimicrob Agents Chemother* 2006;50:2167–2177. [PubMed: 16723579]
5. He H. *Appl Microbiol Biotechnol* 2005;67:444–452. [PubMed: 15702316]
6. Weiss WJ, Murphy T, Lenoy EB, Young M. *Antimicrob Agents Chemother* 2004;48:1708–1712. [PubMed: 15105124]
7. Petersen PJ, Wang TZ, Dushin RG, Bradford PA. *Antimicrob Agents Chemother* 2004;48:739–746. [PubMed: 14982758]
8. Dushin RJ, Wang T-Z, Sum P-E, He H, Sutherland AG, Ashcroft JS, Graziani EI, EKF, Bradford PA, Petersen PJ, Wheless KL, How D, Torres N, Lenoy EB, Weiss WJ, Lang SA, Projan SJ, Shlaes DM, Mansour TS. *J Med Chem* 2004;47:3487–3490. [PubMed: 15214775]
9. Ruzin A, Singh G, Severin A, Yang Y, Dushin RG, Sutherland AG, Minnick A, Greenstein M, May MK, Shlaes DM, Bradford PA. *Antimicrob Agents Chemother* 2004;48:728–738. [PubMed: 14982757]
10. Haltli B, Tan Y, Magarvey NA, Wagenaar M, Yin X, Greenstein M, Hucul JA, Zabriskie TM. *Chem Biol* 2005;12:1163–1168. [PubMed: 16298295]
11. Babu RS, Guppi SR, O'Doherty GA. *Org Lett* 2006;8:1605–1608. [PubMed: 16597121]
12. Adinolfi M, Giacomini D, Iadonisi A, Quintavalla A, Valerio S. *Eur J Org Chem* 2008:2895.
13. Kolb HC, VanNieuwenhze MS, Sharpless KB. *Chem Rev* 1994;94:2483–2547.
14. (a) Fleming PR, Sharpless KB. *J Org Chem* 1991;56:2869–2875. (b) Denis J, Correa A, Greene AE. *J Org Chem* 1990;55:1957–1959. (c) Watson KG, Fung YM, Gredley M, Bird GJ, Jackson WR, Gountzos H, Matthews BR. *J Chem Soc, Chem Commun* 1990:1018.
15. Campbell AD, Raynham TM, Taylor RJK. *Synthesis* 1998:1707–1709.
16. Pitsch S, Weiss PA, Stutz JA, Wu X. *Helv Chim Acta* 2001;84:3773–3795.

Scheme 1.
Synthetic Strategy

Scheme 2.
 Synthesis of Common Intermediate **2**

Scheme 3. Synthesis of the Diastereomeric β -Hydroxyenduracididines from Common Intermediate **2**

Table 1

Structures and Activities of the Mannopeptimycins

mannopeptimycin α - ϵ 1a; β 1b; α $R', R'', R''' = H$ 1c; γ $R' = \text{isovaleroyl}; R'', R''' = H$ 1d; δ $R', R''' = H; R'' = \text{isovaleroyl}$

mannopeptimycin α - ϵ

1e; ϵ R', R'' = R; R''' = isovaleroyl