

## SOLID-PHASE SYNTHESIS OF CYCLIC GLYCOPEPTIDES RELATED TO MANNOPEPTIMYCIN DERIVATIVES

Ting-Zhong Wang,<sup>a\*</sup> Karen L. Wheless,<sup>b</sup> Alan G. Sutherland,<sup>b</sup> and Russell G. Dushin<sup>b</sup>

<sup>a</sup>Chemical Development, <sup>b</sup>Medicinal Chemistry, Chemical and Screening Sciences, Wyeth Research, Pearl River, NY 10965, USA. E-mail: Wangt@wyeth.com

**Abstract** – The mannopeptimycins comprise a novel series of glycopeptide antibiotics that display activity against susceptible and resistant forms of gram-positive bacteria. Low isolated yields of these products from fermentation make the synthesis of easily accessible analogs attractive. A simplified hexapeptide is synthesized using a combination of solid-phase and solution-phase techniques.

Naturally occurring and semisynthetic mannopeptimycins comprise a novel class of glycopeptide antibiotics that display activities against both susceptible and resistant forms of gram-positive bacteria.<sup>1-3</sup> The key structural feature is a cyclic hexapeptide core that contains alternating D- and L-amino acids and includes an epimeric pair of a previously unknown amino acid,  $\beta$ -hydroxyenduricididine. One of these

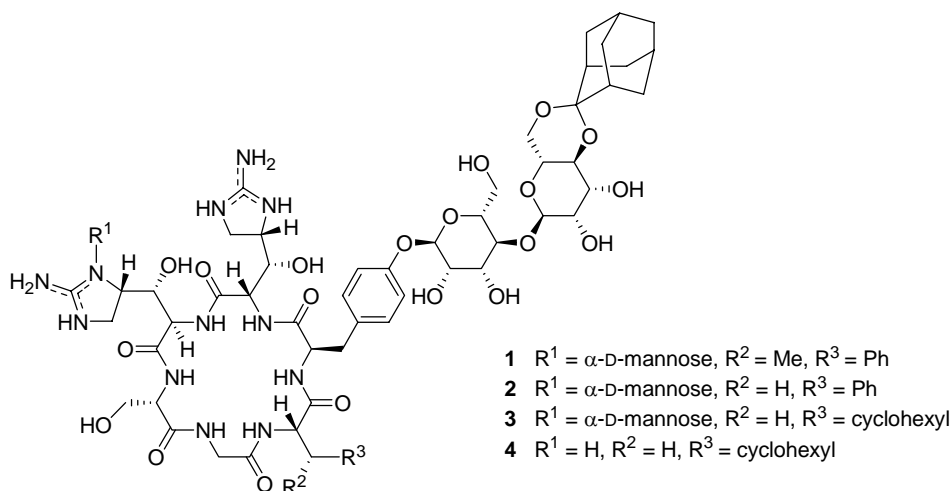
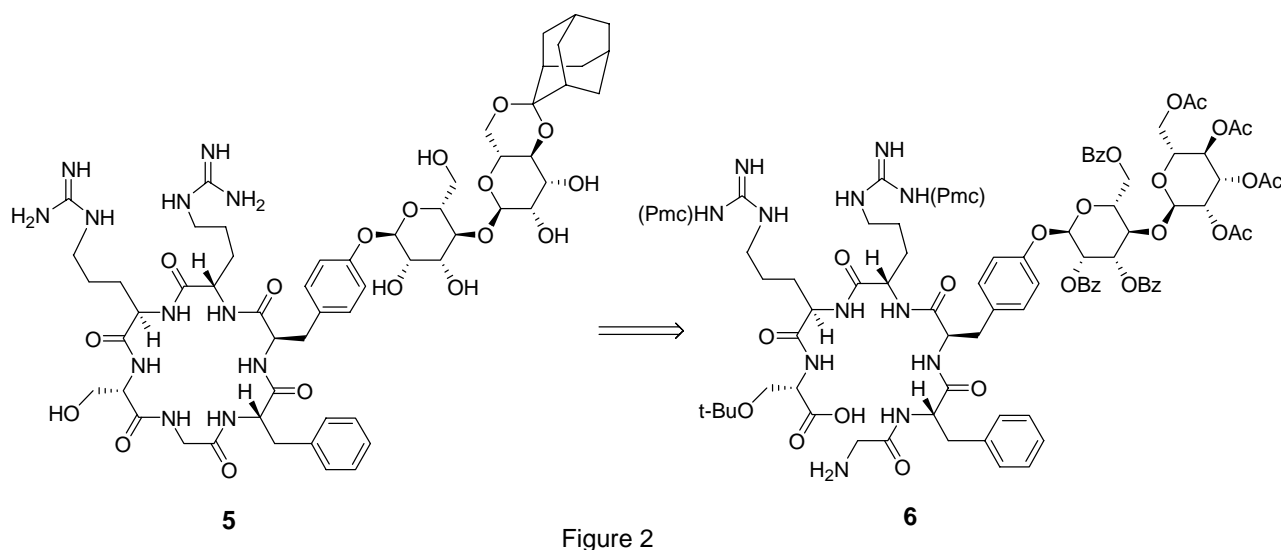


Figure 1

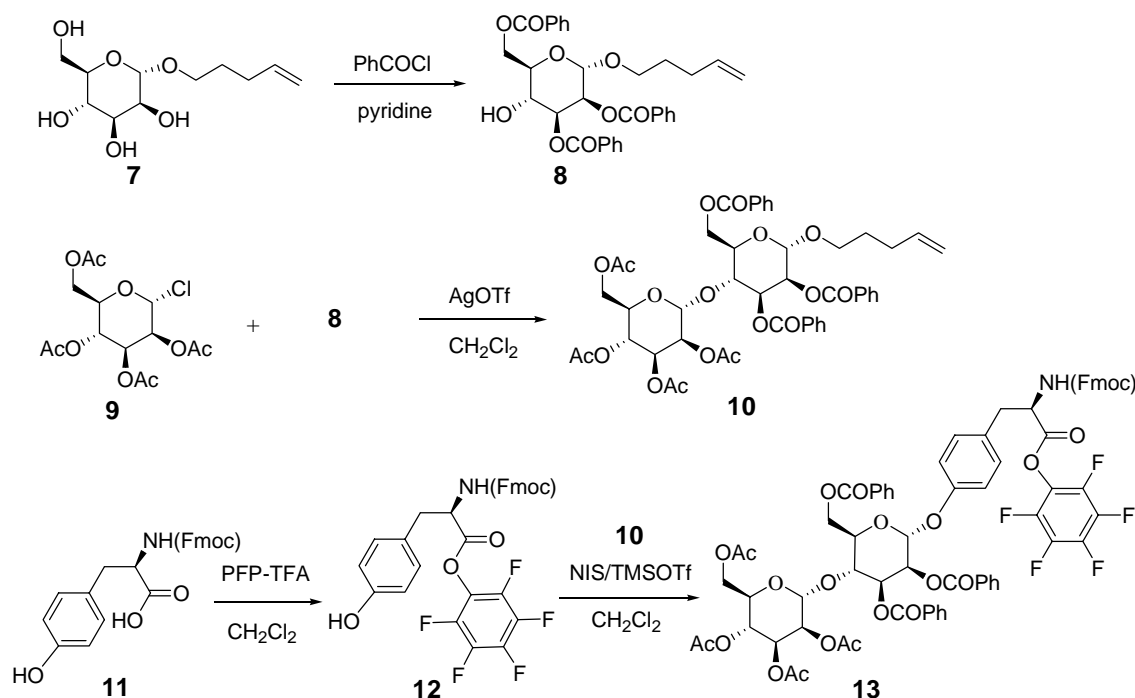
novel residues contains a mannopyranose moiety in the  $\alpha$ -configuration while a D-tyrosine residue contains an  $\alpha$ -linked mannose disaccharide. In the natural series, isovalerate substitution occurs on the terminal mannose of this disaccharide, and this substitution conveys substantial increases in antibacterial potency over the non-derivatized parent molecule.<sup>2</sup> Similarly, those semisynthetic variants that display potent antibacterial activities are derivatized on the same terminal mannose.<sup>3</sup> Notably, replacements of the  $\beta$ -methylphenylalanine residue with phenylalanine or cyclohexylalanine are tolerated, and derivatives lacking the *N*-linked mannose or the hydroxyl substituents on the  $\beta$ -hydroxyenduricididine side chains also retain good levels of antibacterial activities. Among the more potent and most easily accessed semisynthetic derivatives are adamantyl ketals (**1-4**) (Figure 1).<sup>3</sup> The apparent tolerability of structural variation on these three residues provided impetus to prepare simplified derivatives in which the  $\beta$ -hydroxyenduricididine residues are replaced with arginines. Herein we report results on the total synthesis of a representative compound.

Retrosynthetically, we envisioned that target compound (**5**) could be derived from an appropriately protected linear glycopeptide (**6**) which could be assembled using solid-phase techniques (Figure 2). We anticipated that cyclization to provide the hexapeptide core would be most readily accomplished with amide bond formation between the L-serine and unhindered glycine residues. Five of the six requisite amino acids are commercially available, while the D-tyrosine containing a mannose disaccharide would be prepared synthetically.

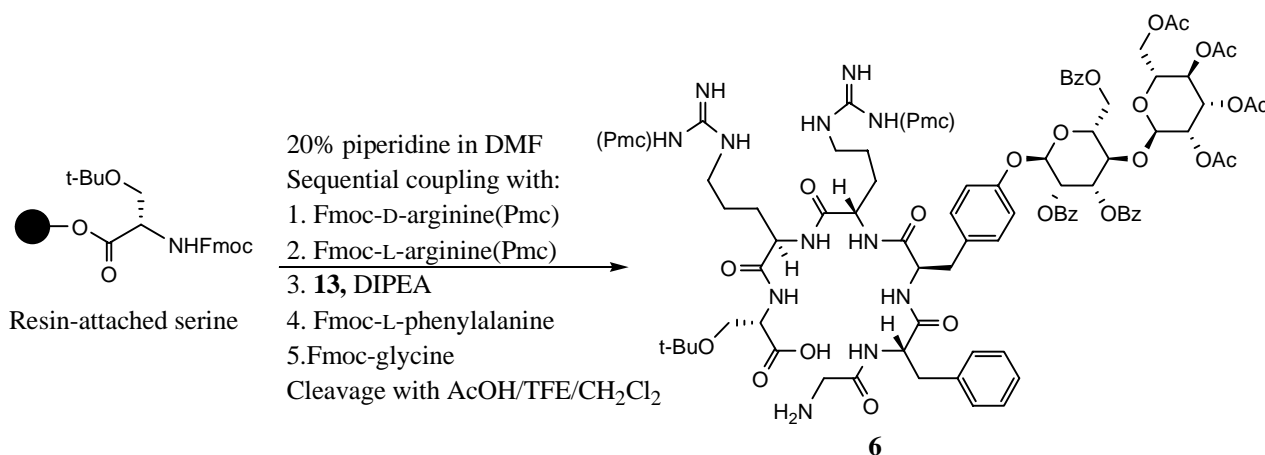


Synthesis of the disaccharide started with the known pentenyl- $\alpha$ -D-mannopyranoside (**7**)<sup>4</sup> which was selectively protected with benzoyl chloride to give pentenyl-2,3,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranoside (**8**) retaining a hydroxy substituent on the 4-position open for glycosylation<sup>5</sup> (Scheme 1). Dimannoside (**10**) was obtained in modest yield by silver(I) triflate-promoted coupling of **8** with 1- $\alpha$ -chlorotetraacetylmannose (**9**).<sup>6</sup> Prior to glycosylation, the commercially available *N*-Fmoc-tyrosine

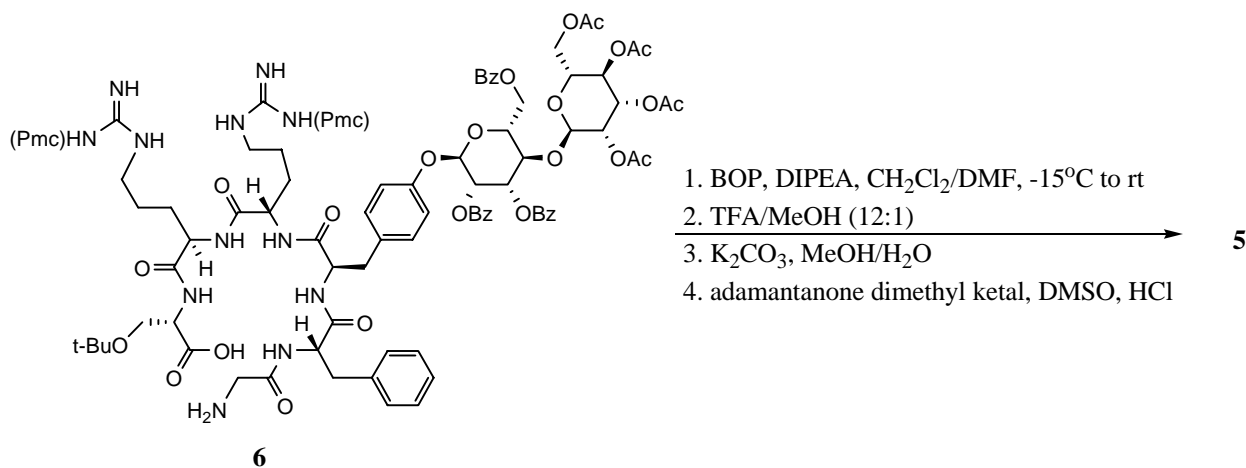
(**11**) was reacted with pentafluorophenyl trifluoroacetate (PFPTFA) to give pentafluorophenyl ester (**12**) which is suitably protected for glycosylation and activated for amide bond formation during peptide synthesis.<sup>7</sup> Coupling of the dimannoside (**10**) with **12** was mediated by *N*-iodosuccinimide/trimethylsilyl triflate and gave the glycosylated tyrosine derivative (**13**) required for peptide synthesis.<sup>4</sup> The stereochemistry of both  $\alpha$ -anomeric centers was assigned based on literature precedents<sup>4,6</sup> and was confirmed by <sup>1</sup>H and <sup>13</sup>C NMR spectroscopy.<sup>8</sup>



For solid-phase synthesis, we employed a 2-chlorotrityl chloride resin with HBTU/HOBt coupling protocols and Fmoc protection. The synthesis commenced with attachment of *O*-*tert*-butyl-*N*-Fmoc-serine to the resin. The loading of the resin was determined to be in the range of ~0.5 mmol/g by UV quantification of the Fmoc-piperidine adduct.<sup>9</sup> The linear peptide chain was then assembled on the resin



in a sequential fashion using standard procedures (Scheme 2). Peptide coupling reactions were monitored for completion by negative Kaiser ninhydrin test. Once peptide elongation was accomplished, the protected linear hexapeptide (**6**) was released from the resin by cleavage with a mixture of acetic acid, trifluoroethanol and dichloromethane (1:1:8). The cyclization of **6** was effected by using BOP/DIPEA as the dehydrating reagent in a mixed solvent system of DMF/CH<sub>2</sub>Cl<sub>2</sub> (1:1). The cyclic peptide was thereby obtained cleanly with almost complete suppression of dimerization when the concentration of the substrate was held at <math>10^{-2}</math> M. Deprotection was accomplished sequentially by treatment with TFA/MeOH to remove the *tert*-butyl and the 2,2,5,7,8-pentamethylchroman-6-sulfonyl (Pmc) groups, and then with K<sub>2</sub>CO<sub>3</sub>/MeOH for hydrolysis of the acetates and benzoates. The fully deprotected glycopeptide was converted to the final target (**5**) by transketalization with adamantane dimethyl ketal (Scheme 3). The selectivity at 4,6-position of the terminal mannose is in accordance to those observed for natural mannopeptimycins with the formation of only a small amount of the corresponding 2,3-ketal isomer. To our surprise, the synthetic glycopeptide (**5**) was found to have very poor activity against a diverse panel of bacteria. The disparity of activities between this fully synthetic analog and the semisynthetic derivatives (**1-4**) demonstrates that the cyclic guanidines on the  $\beta$ -hydroxyenduricidine residues play a very important role in conveying antibacterial activities of the mannopeptimycin derivatives.



Scheme 3

## ACKNOWLEDGEMENTS

The authors gratefully acknowledge the support of Magid Abou-Gharbia, Tarek Mansour, and Janis Upeslakis. Xidong Feng, Kaiko Tabei, Murthy Damarla, Rachel Garlish, Mark Tischler and Joseph Ashcroft are acknowledged for providing separations and analytical support.

## REFERENCES AND NOTES

1. S. E. De Voe and M. P. Kunstmann, "Antibiotic AC98 and Production Thereof." US Patent 3,495,004, Feb. 10, 1970 (*Chem. Abstr.*, 1970, **72**, 223).

2. H. He, R. T. Williamson, B. Shen, E. I. Graziani, H. Y. Yang, S. M. Sakya, P. J. Petersen, and G. T. Carter, *J. Am. Chem. Soc.*, 2002, **124**, 9729.
3. P. E. Sum, D. How, N. Torres, P. J. Petersen, E. B. Lenoy, W. J. Weiss, and T. S. Mansour, *Bioorg. Med. Chem. Lett.*, 2003, **13**, 1151; P. E. Sum, D. How, N. Torres, H. Newman, P. J. Petersen, and T. S. Mansour, *Bioorg. Med. Chem. Lett.*, in press; P. E. Sum, D. How, N. Torres, P. J. Petersen, J. Ashcroft, E. I. Graziani, F. E. Koehn, and T. S. Mansour, *Bioorg. Med. Chem. Lett.*, in press; H. He, B. Shen, P. J. Petersen, W. J. Weiss, H. Y. Yang, T. Z. Wang, R. G. Dushin, and G. T. Carter, *Bioorg. Med. Chem. Lett.* (submitted); T. Z. Wang and R. G. Dushin, unpublished results.
4. D. R. Mootoo, P. Konradsson, and B. Fraser-Reid, *J. Am. Chem. Soc.*, 1989, **111**, 8540.
5. J. M. Williams and A. C. Richardson, *Tetrahedron*, 1967, **23**, 1369.
6. S. Akhtar, A. Routledge, R. Patel, and J. M. Gardiner, *Tetrahedron Lett.*, 1995, **36**, 7333.
7. K. J. Jensen, M. Meldal, and K. Bock, *J. Chem. Soc., Perkin Trans. I*, 1993, 2119.
8. For compound (**10**): at both anomeric centers,  $^1J_{\text{CH}} = 172$  Hz, a good indication of  $\alpha$ -configurations.
9. J. Green and K. Bradley, *Tetrahedron*, 1993, **49**, 4141.