Vancomycin CD and DE Macrocyclization and Atropisomerism Studies

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Continued studies on the synthesis and atropisomerism of the vancomycin CD and DE ring systems based on aromatic nucleophilic substitution macrocyclization reactions for formation of the biaryl ethers are detailed in efforts that further define substituent effects, explore the impact of protecting groups, and establish the stereochemical integrity of peripheral substituents. These have led to the identification of a previously unrecognized site of epimerization within our original approach to the DE ring system and the introduction of significant improvements in the approach that will find utilization in syntheses of the vancomycin CDE ring system and of the natural product itself. The preparation of a complete set of DE ring system isomers bearing the unnatural stereochemistry at the labile C8, C11, and C14 sites was accomplished for comparison and established that C8 is prone to epimerization to the more stable, unnatural *S* versus *R* absolute stereochemistry if it bears an ester, but not a carboxamide, substituent. Additionally, an improved synthesis of the CD ring system, enlisting a C14 carboxamide versus ester substituent, is disclosed and establishes the stereochemical integrity of our prior approach which incorporated a C14 ester. A set of fully functionalized CD and DE ring systems were prepared and include the development of conditions for the final deprotections required for incorporation into efforts on the natural product. The examination of the antimicrobial activity of these key substructures of vancomycin is detailed.

Vancomycin (**1**, Figure 1), which was isolated in 1956 from *Streptomyces orientalis*[1] and whose structure was secured 25 years later,[2] is the prototypical member of a class of clinically important glycopeptide antibiotics.[3] It is the therapeutic agent of choice for the treatment of methicillin-resistant *Staphylococcus aureus* and is routinely used against enterococci and bacterial infections in patients allergic to β-lactam antibiotics.[4] The structural complexity of vancomycin, the interest in defining structural features it possesses that are responsible for inhibition of cell-wall biosynthesis in sensitive bacteria,[5] and the emergence of clinical resistance[6] have provided renewed interest in the total synthesis of **1** and related agents.[7−14] In recent studies, we disclosed the synthesis of appropriately functionalized vancomycin CD, DE, and CDE ring systems on the basis of aromatic nucleophilic substitution macrocyclization reactions with formation of the biaryl ethers and detailed an indirect solution to the control of the atropisomer stereochemistry, enlisting a selective DE versus CD atropisomer isomerization.[9−11] Herein, we provide full details of studies that further define the scope of the approach to the DE and CD ring systems which were conducted in an effort to optimize conditions for cyclization, to examine the effect of different protecting groups, to confirm and establish the stereochemistry, and to define thermal atropisomerism substituent effects. These studies have resulted in significant improvements in the approach and have identi-

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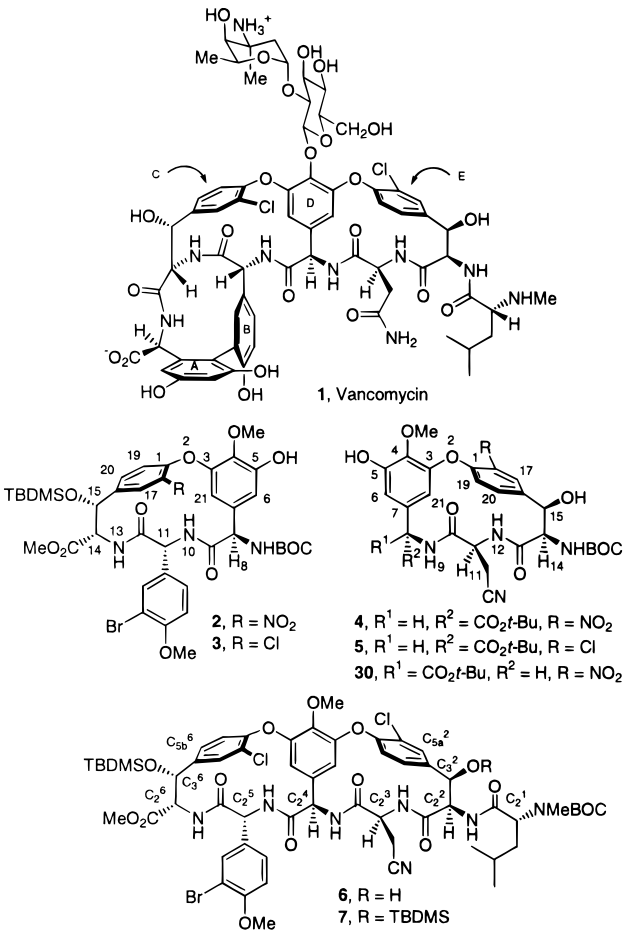


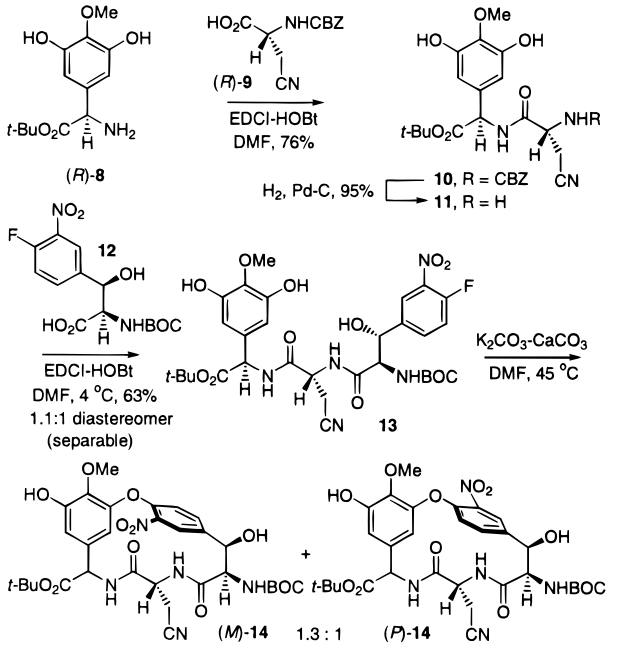
Figure 1.

fied a previously unrecognized site of epimerization[15] within our prior approach to the DE ring system. Since the DE ring system of vancomycin incorporates four of the five key H-bonding sites for NAc-D-Ala-D-Ala binding, the biological evaluation of a set of fully functionalized DE and CD ring systems was conducted and constitutes the examination of key substructures of the natural product.

C11 Diastereomer of the DE Ring System. In the course of efforts on the preparation of the DE ring system, the C11 and C14 centers emerged as sites that are susceptible to epimerization when activated for coupling at the adjacent carboxylate. Typically, the couplings that link C13 to N12 or enlist the N-terminus dipeptide when constructing C10−N9 require effort to minimize racemization, suggesting that substrates incorporating the dipeptide may be prone to epimerization. To ensure that such an epimerization at either site does not accompany macrocyclization, both **14** and **16** were prepared for comparison.

The preparation of **14** is detailed in Scheme 1. Thus, coupling of *N*-CBZ-(*R*)-(*β*-cyano)alanine (**9**)[16] with *tert*-butyl (*R*)-(3,5-dihydroxy-4-methoxyphenyl)glycine (**8**, ≥99% ee)[11,12] provided **10** as a single detectable diastereomer (76%). *N*-CBZ deprotection (H_2, Pd/C, 95%) followed by coupling with *N*-BOC-3-(2*R*,3*R*)-hydroxy-3-(4-fluoro-3-nitrophenyl)alanine[11,15c] (**12**) provided **13** accompanied by a significant amount of the diastereomer derived from C14 epimerization.[17] Without optimization, subjection of **13** to macrocyclization by treatment with K_2CO_3/$CaCO_3$ (5.0 equiv/7.5 equiv, 2 wt equiv of 4 Å MS, 0.008 M DMF, 45 °C, 49 h) provided a separable 1.3:1 mixture of the two atropisomers of **14** with the unnatural *M* atropisomer predominating slightly.[18] Although not examined in detail, the ring closure appears slower and lower yielding than that used to prepare **4**.[11] The comparison of (*M*)- and (*P*)-**14** with both atropisomers of **4** and the minor C8 diastereomer[19] of **4** confirmed they were distinct and that the macrocyclization enlisted in the preparation of **4** proceeded without detectable C11 epimerization.[20]

Scheme 1



C14 Diastereomer of the DE Ring System. During the course of prior studies, ample quantities of the minor diastereomer **15**,[11] derived from coupling of *tert*-butyl (*S*)-(*β*-cyano)alanyl-(*R*)-(3,5-dihydroxy-4-methoxyphenyl)gly-

(15) For examples of such epimerizations preceding or accompanying macrocyclization with biaryl ether formation: (a) Pearson, A. J.; Zhang, P.; Bignan, G. *J. Org. Chem.* **1997**, *62*, 4536. (b) Boger, D. L.; Zhou, J. *J. Org. Chem.* **1996**, *61*, 3938. Boger, D. L.; Zhou, J.; Borzilleri, R. M.; Nukui, S. *Bioorg. Med. Chem. Lett.* **1996**, *6*, 1089. (c) Boger, D. L.; Zhou, J.; Borzilleri, R. M.; Nukui, S.; Castle, S. L. *J. Org. Chem.* **1997**, *62*, 2054. (d) For observations analogous to those described herein, see: Pearson, A. J.; Chelliah, M. V. *J. Org. Chem.* **1998**, *63*, 3087.

(16) *N*-CBZ-D-(*β*-cyano)alanine (**9**) was prepared following the procedure detailed for the L enantiomer: CBZNH-D-Asn (1.0 equiv DCC, pyridine, 55%), Ressler, C.; Ratzkin, H. *J. Org. Chem.* **1961**, *26*, 3356. For D-**9**: $[\alpha]^{25}_D$ +43 (*c* 0.5, DMF). (*S*)-**8** was prepared by enlisting the $(DHQ)_2PHAL$ ligand in the AA reaction following the procedure detailed[11,12] for the *R* enantiomer. For (*S*)-**8**: $[\alpha]^{25}_D$ +54 (*c* 0.27, CH_3-OH). (*S*)-**41**: $[\alpha]^{25}_D$ +74 (*c* 0.8, CH_3OH), was prepared as an intermediate en route to (*S*)-**8** and converted to **43**: $[\alpha]^{25}_D$ +13 (*c* 0.08, $CHCl_3$) EDCI−HOAt, CH_2Cl_2-DMF 4:1, 4 °C, 18 h, 85%, $[\alpha]^{25}_D$ +142 (*c* 0.4, $CHCl_3$); H_2, Pd/C, CH_3OH−EtOAc 1:1, 25 °C, 5 h, 98%.

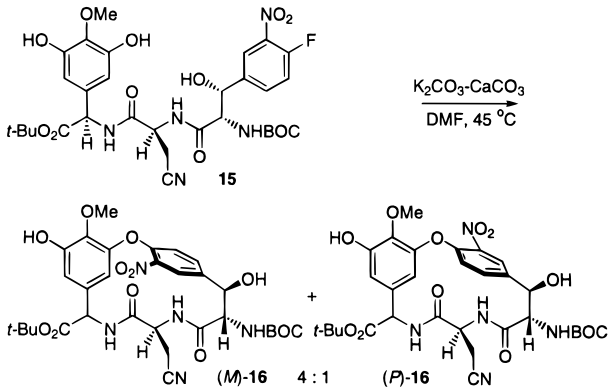
(17) Upon completion of this work, we have established that the predominant extent of C14 epimerization occurs upon hydrolysis of the precursor methyl ester rather than during the amide coupling reaction. This can be minimized by conducting the hydrolysis at 0 °C for a minimum reaction period.

(18) In preceding reports, we believe we have reversed the *M* and *P* stereochemical representations which may be attributed to ambiguities in the assignment of the pilot atom and its orientation for assignment of *M* and *P*: Eliel, E. L.; Wilen, S. H. *Stereochemistry of Organic Compounds*; Wiley: New York, 1994; pp 1120−1122 versus Cahn, R. S.; Ingold, C.; Prelog, V. *Angew. Chem., Int. Ed. Engl.* **1966**, *5*, 385. For the present assignments, the pilot atoms selected are the C15 OR (CD) and C15 OR (DE) substituents, and the chiral plane (the C ring for CD, the E ring for DE) is viewed from the point of view of the pilot atom (i.e., the pilot atom is placed above the chiral plane).

cine with **12**, were accumulated. Without optimization, subjection of **15** to macrocyclization with $K_2CO_3/CaCO_3$ (5.0 equiv/7.5 equiv, 2 wt equiv of 4 Å MS, 0.008 M DMF, 48 °C, 24 h) provided a 4:1 mixture of the two separable atropisomers of **16** (43%) in which the unnatural *M*-isomer was favored (Scheme 2), and the closure proceeded at a rate analogous to that observed with **4**. The comparison of **4** and its minor C8 diastereomer[19] with the two atropisomers of **16** confirmed that they are distinguishable and that the preparations of **4** proceeded without detectable C14 epimerization.[20] Attempts to cyclize **15** with CsF (5 equiv, 0.005 M DMF or DMSO, 25 °C) provided a much more polar set of atropisomers in yields as high as 75% containing isomers which were easily distinguishable from **4** or **16**. These possessed a higher molecular weight, indicative of further substrate reactions, and although their identification was not pursued, more detailed studies with **4** implicated C8 oxidation.[21]

Alternative L-Asn Residues. In our prior studies, the L-Asn residue in the DE ring system was protected

Scheme 2



enlisting the corresponding nitrile. A (4,4'-dimethoxydiphenyl)methyl (Ddm)[22] protected L-Asn was also examined in light of its use in related efforts[8,14] to assess whether it might favorably impact the macrocyclization reaction. In particular, we were interested in establishing whether the liberated fluoride might prove sufficiently basic to promote β-elimination through α-deprotonation of the nitrile, thus lowering the apparent effectiveness of our original approach. Thus, coupling of *N*-CBZ-*N'*-Ddm-(*S*)-Asn[22] (**17**) with (*R*)-**8** cleanly provided the dipeptide **18** (92%) as a single detectable diastereomer (Scheme 3). *N*-CBZ deprotection (H_2, Pd/C, 100%) followed by coupling of the free amine **19** with **12** (2.2 equiv of EDCI, 1.2 equiv of HOBt, DMF, 0−4 °C, 15 h, 73%) provided the tripeptide **20** as a separable 1.8:1 mixture of diastereomers without optimization.[17] Under comparable conditions, the L-(β-cyano)alanine-containing dipeptide provided a more favorable 6:1 ratio of C14 diastereomers.[11] Macrocyclization upon treatment with $K_2CO_3/CaCO_3$ (5.0 equiv/7.7 equiv, 2 wt equiv of 4 Å MS, 0.008 M DMF, 45 °C, 47 h) provided a mixture of the two atropisomers of **21**[20] in a yield (47% versus 59%) and atropisomer diastereoselectivity (1:1.3 versus 1:1.3 *M*:*P*) comparable, but not superior, to the substrate bearing a nitrile. Analogous to observations made with **16** and in our prior efforts,[11] attempted closure of **20**, enlisting CsF

(19) Trace amounts (<7−8%) of a C8 diastereomer were detected and isolated when the macrocyclization leading to **4** was prematurely quenched. The diastereomer, not detected in the closure of **23**, rapidly and completely epimerized to **24** upon treatment with base. Thus, treatment of (*P*)-**30** or (*P*)-**24** with 2 equiv of DBU (THF-d_8, 25 °C) or 3 equiv of K_2CO_3 (DMF-d_7, 25 °C) provided a 3−5:95−97 mixture of (*P*)-**30**:(*P*)-**24** in the time that it takes to immediately record the ^{1}H NMR. Conformational searches conducted on both atropisomers of the 8*R* and 8*S* diastereomers **30**/**24**, **28**/**29**, **37**/**45** (MacroModel BatchMin 6.0, OPLSA Force Field) revealed a preference for the unnatural 8*S* diastereomer (ΔE = 2.3−3.9 kcal/mol) which is consistent with the experimental observations, the epimerization of **30** to **24**, and the assigned stereochemistries. The natural 8*R*-**37** proved more resistant toward C8 epimerization than **30**. Thus, treatment of **37** with 2 equiv of DBU (THF, 25 °C, 40 min), 5 equiv of K_2CO_3 (DMF, 45 °C, 12 h), or 5 equiv CsF (DMF, 25 °, 8 h) led to generation of 20%, 58%, and 5%, respectively, of the C8 diastereomer **45** and the equilibrium ratio of 8*S*:8*R* was established to be 95:5. Data for *tert*-butyl (*M*)- and (*P*)-(8*R*,-11*S*,14*R*,15*R*)-14-[*N*-[(*tert*-butyloxy)carbonyl]amino]-11-(cyanomethyl)-5, 15,-dihydroxy-10,13-dioxo-4-methoxy-18-nitro-9,12-diaza-2-oxatricyclo-[14.2.2.1^{3,7}]heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxylate (**30**) follow. (*M*)-**30** (the less polar isomer): $[\alpha]^{25}_D$ +78 (*c* 0.22, CH_3OH); ^{1}H NMR (acetone-d_6, 400 MHz) δ 8.28−8.21 (m, 1H), 8.20 (s, 1H), 8.16 (s, 1H), 7.96 (d, 1H, *J* = 8.1 Hz), 7.53−7.47 (m, 1H), 7.44 (d, 1H, *J* = 8.4 Hz), 6.60 (s, 1H), 6.27 (d, 1H, *J* = 8.4 Hz), 5.46 (d, 1H, *J* = 1.9 Hz), 5.27 (s, 1H), 5.14−5.06 (m, 1H), 4.99 (d, 1H, *J* = 7.0 Hz), 4.73−4.68 (m, 1H), 4.62−4.55 (m, 1H), 3.95 (s, 3H), 2.93−2.77 (m, 2H), 1.44 (s, 9H), 1.40 (s, 9H); IR (neat) ν_{max} 3326, 1660, 1537, 1341, 1249, 1158 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 804.1529 (M^+ + Cs, $C_{31}H_{37}N_5O_{12}$ requires 804.1493). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 25 °C) of (*M*)-**30** exhibited the following diagnostic NOE cross-peaks: H-20/H-19 (s, δ 7.75/7.35), H-20/H-15 (s, δ 7.75/5.15), H-20/H-14 (s, δ 7.75/4.56), H-15/H-14 (s, δ 5.15/4.56), H-6/H-8 (m, δ 6.50/4.77), H-11/CH_2CN (m, δ 4.48/2.84 and 4.48/2.71). For (*P*)-**30** (the more polar isomer): $[\alpha]^{25}_D$ +65 (*c* 0.18, CH_3OH); ^{1}H NMR (acetone-d_6, 400 MHz) δ 8.25 (s, 1H), 8.20 (s, 1H), 7.94 (d, 1H, *J* = 5.6 Hz), 7.82 (d, 1H, *J* = 7.3 Hz), 7.76 (d, 1H, *J* = 8.4 Hz), 7.26 (d, 1H, *J* = 8.6 Hz), 6.66 (s, 1H), 6.34 (d, 1H, *J* = 7.6 Hz), 5.69 (s, 1H), 5.31−5.26 (m, 1H), 5.13 (d, 1H, *J* = 7.8 Hz), 5.07 (d, 1H, *J* = 6.8 Hz), 4.68−4.60 (m, 1H), 4.56 (d, 1H, *J* = 5.6 Hz), 3.94 (s, 3H), 2.85−2.75 (m, 2H), 1.44 (s, 9H), 1.38 (s, 9H); IR (neat) ν_{max} 3410, 3287, 1668, 1634, 1537, 1254, 1158 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 804.1529 (M^+ + Cs, $C_{31}H_{37}N_5O_{12}$ requires 804.1493). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 25 °C) of (*P*)-**30** exhibited the following diagnostic NOE cross-peaks: H-17/H-15 (s, δ 8.12/5.14), H-17/H-14 (s, δ 8.12/4.41), H-20/H-19 (s, δ 7.79/7.17), H-19/H-21 (m, δ 7.17/5.62), H-15/H-14 (s, δ 5.14/4.41), H-6/H-8 (s, δ 6.53/4.79), H-11/CH_2CN (m, δ 4.52/2.78 and 4.52/2.70).

(20) Tentative C8 stereochemical assignments for **16** and **21** are also the unnatural, epimerized 8*S* stereochemistry. A conformational search of both atropisomers of the C8 diastereomers of **16** (MacroModel BatchMin 6.0, OPLSA) established that the unnatural 8*S* diastereomers were substantially more stable (ΔE = >2.5 kcal/mol). For **21**, this is analogous to observations made with **4** and further supported by its relative ease of atropisomerism, see Table 2. Product **14** has been tentatively assigned the natural 8*R* stereochemistry on the basis of a conformational search (MacroModel, BatchMin 6.0, OPLSA) conducted on both atropisomers of the C8 diastereomers in which the natural, nonepimerized 8*R* configuration was established to be more stable (ΔE = 1.1−2.7 kcal/mol).

(21) The chemical shifts of C6-H, C21-H, C8-H, and N9-H shift dramatically (>1 ppm) and the signals previously attributable to C8-H and N9-H collapse to singlets versus doublets. For the major atropisomer derived from **15**: $[\alpha]^{25}_D$ −11.6 (*c* 0.01, $CHCl_3$); ^{1}H NMR (acetone-d_6, 400 MHz) δ 9.22 (s, 1H), 8.21 (d, 1H, *J* = 8.2 Hz), 8.11 (s, 1H), 7.76 (d, 1H, *J* = 8.7, 2.2 Hz), 7.36 (d, 1H, *J* = 2.1 Hz), 7.26 (br s, 1H), 7.13 (s, 1H), 7.11 (d, 1H, *J* = 7.2 Hz), 6.76 (br s, 1H), 6.47 (d, 1H, *J* = 8.3 Hz), 5.47 (d, 1H, *J* = 4.2 Hz), 5.11−5.09 (m, 1H), 4.77−4.71 (m, 1H), 4.35 (t, 1H, *J* = 8.5 Hz), 3.92 (s, 3H), 3.05 (dd, 1H, *J* = 17.1, 5.3 Hz), 2.93 (dd, 1H, *J* = 17.1, 7.6 Hz), 1.56 (s, 9H), 1.30 (s, 9H); IR (film) ν_{max} 3313, 2923, 1676, 1539, 1349, 1246, 1133 cm^{-1}; FABMS (NBA-NaI) *m*/*z* 710 ($C_{31}H_{37}N_5O_{12}$ requires 694). For the product derived from **20** (40%): ^{1}H NMR (acetone-d_6, 400 MHz) δ 9.09 (s, 1H), 8.18−8.14 (m, 2H), 7.97 (d, 1H, *J* = 8.0 Hz), 7.77 (dd, 1H, *J* = 8.8, 2.1 Hz), 7.35 (d, 1H, *J* = 2.0 Hz), 7.28 (s, 1H), 7.19−7.16 (m, 4H), 7.12−7.08 (m, 2H), 6.88−6.82 (m, 4H), 6.55 (s, 1H), 6.22 (d, 1H, *J* = 9.2 Hz), 6.10 (d, 1H, *J* = 8.3 Hz), 5.80 (d, 1H, *J* = 3.5 Hz), 4.94−4.90 (m, 1H), 4.78−4.74 (m, 1H), 4.33 (t, *J* = 8.6 Hz), 3.86 (s, 3H), 3.76 (s, 3H), 3.75 (s, 3H), 1.54 (s, 9H), 1.23 (s, 9H); FABMS (NBA-NaI) *m*/*z* 955 (M^+ + Na, $C_{46}H_{53}O_{15}N_5$ requires 939). For the products derived from **31** (70%): ^{1}H NMR (acetone-d_6, 400 MHz) δ 9.09 (s, 1H), 8.19 (d, 1H, *J* = 7.8 Hz), 8.14 (s, 1H), 7.77 (dd, 1H, *J* = 8.5, 2.2 Hz), 7.35 (d, 1H, *J* = 2.0 Hz), 7.35−7.33 (m, 1H), 7.12 (s, 1H), 7.11 (d, 1H, *J* = 8.6 Hz), 6.78−6.76 (m, 1H), 6.31 (d, 1H, *J* = 9.0 Hz), 5.61 (d, 1H, *J* = 3.1 Hz), 5.01 (d, 1H, *J* = 9.0 Hz), 4.80−4.76 (m, 1H), 4.43 (t, 1H, *J* = 9.0 Hz), 3.92 (s, 3H), 2.98−2.82 (m, 2H), 1.56 (s, 9H), 1.18 (s, 9H); IR (film) ν_{max} 3313, 2923, 2246, 1676, 1539, 1349, 1133 cm^{-1}; FABMS (NBA-NaI) *m*/*z* 710 (M^+ + Na, $C_{31}H_{37}O_{12}N_5$ requires 694).

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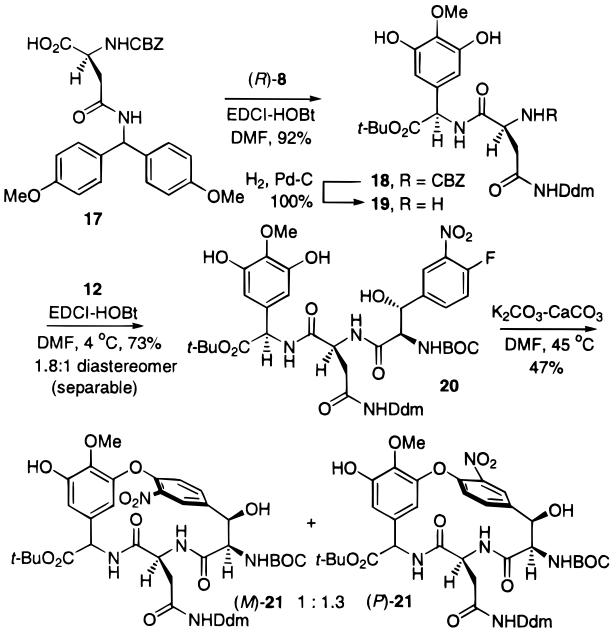
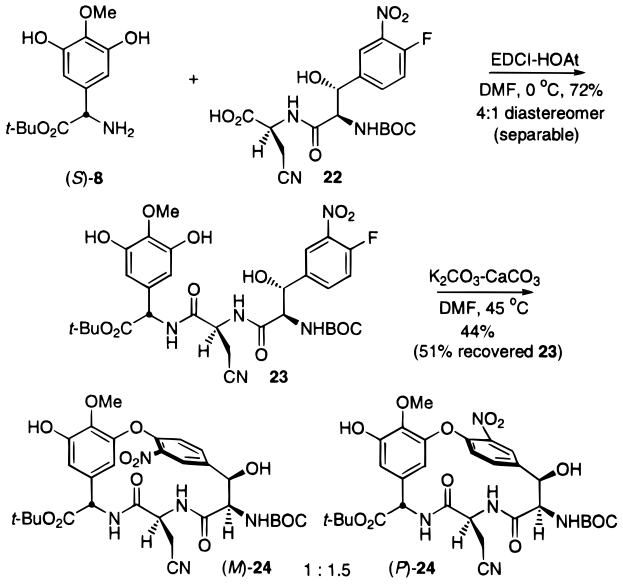
Scheme 3



Scheme 4



(5 equiv, 25 °C, 0.008 M) in either DMF (48 h) or DMSO (24 h), provided only low yields of **21** (21%, 1:1 mixture of atropisomers) and afforded, predominately, a much more polar cyclized product (40%)[21] that possessed a molecular weight 16 mass units higher than the desired products. Although the identity of this undesired cyclization product was not established, analysis of the ^{1}H NMR indicated potential epimerization and oxidation of the C8 center under the reaction conditions.[21] Important for our considerations, this established that the failure of the CsF-promoted closure with **4**[11] was not attributable to a fluoride-induced β-elimination within the nitrile-containing L-Asn subunit. Unlike samples of **4**, a sample of **21** was only partially resolved by chromatography (3.5% $CH_3OH-CHCl_3$, PTLC) into a 1.8:1 mixture enriched in the less polar *M* atropisomer and a 1:3.2 mixture enriched in the more polar *P* atropisomer. This difficulty in separating the two atropisomers coupled with the slightly lower macrocyclization conversions and the failure to withstand CsF-promoted closure suggests no immediate advantage to the use of **20** and a Ddm-protected Asn.

C8 Diastereomer of the DE Ring System. The acidic C8 center on the central phenylglycine had been anticipated to be problematic, and our efforts to promote the closure to **4** with CsF versus K_2CO_3 were not successful[11] and provided products implicating additional reactions at this center. This behavior required the preparation and comparison of the C8 diastereomer **24** in order to unambiguously establish the expected stereochemical assignments. Thus, coupling of **22**[11] with (*S*)-**8**[16] provided **23** (72%) accompanied by a separable minor diastereomer derived from C11 epimerization (Scheme 4). Macrocyclization of **23** upon treatment with K_2CO_3/$CaCO_3$ (5.0 equiv/7.5 equiv, 2 wt equiv of 4 Å MS, 0.008 M DMF, 45 °C, 35 h) cleanly provided a 1:1.5 mixture of the separable atropisomers of **24** (44% unoptimized) whose atropisomer stereochemistries were established by 2D ^{1}H−^{1}H NMR along with recovered starting material (51%). Ultimately, the putative C8 diastereomer **24** proved identical to our prior samples of **4**, indicating C8 epimerization to provide a single dominant, albeit unassigned, C8 diastereomer.[19] This comparison was initially complicated by what proved to be a concentration-dependent ^{1}H NMR behavior attributable now to its dimerization or oligomerization in relatively nonpolar solvents.[15] Conformational analyses[19] of **4** and **24** revealed a significant preference for the unnatural 8*S* diastereomer, consistent with an unrecognized epimerization that accompanied cyclization to provide **4**. As outlined below, this was further implicated in deliberate epimerization studies of a minor diastereomer isolated along with **4** and verified by the correlations of **24**, our prior samples of **4**, and its minor diastereomer[19] with the C8 hydroxymethyl and *N*-methyl carboxamide cyclization products **28** and **37**/**45**, which were established to close without significant competitive epimerization.

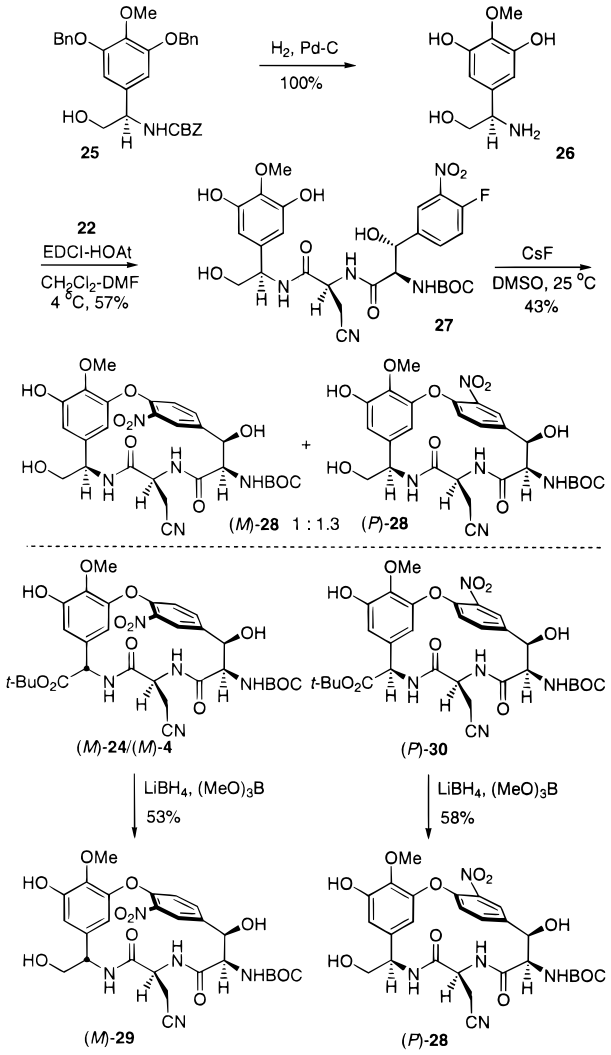
Establishment of the C8 Stereochemistry. The C8 stereochemistry was unambiguously established by examining the (8*R*)-hydroxymethyl derivative **28**, incapable of epimerization upon cyclization of **27** (Scheme 5), and its correlation with the reduction products derived from **24** and with its C8 diastereomer.[19] *N*-CBZ and benzyl ether deprotection of **25**[11,12] (H_2, Pd/C, 100%) followed by coupling of the free amine **26** with **22**[11] provided **27** (57%). Macrocyclization of **27** upon treatment with CsF (DMSO, 25 °C, 24 h) provided **28** as a separable 1.3:1 mixture of atropisomers (Scheme 5).

$LiBH_4$ reduction[23] of samples of (*M*)-**4** which were identical with (*M*)-**24** did not provide (*M*)-**28** establishing their C8 stereochemistry as the unnatural 8*S* configuration. In contrast, the minor C8 diastereomer[19] (*P*)-**30** did provide (*P*)-**28** upon reduction, establishing its 8*R* stereochemistry and the strong thermodynamic preference for the unnatural 8*S* stereochemistry (Scheme 5).

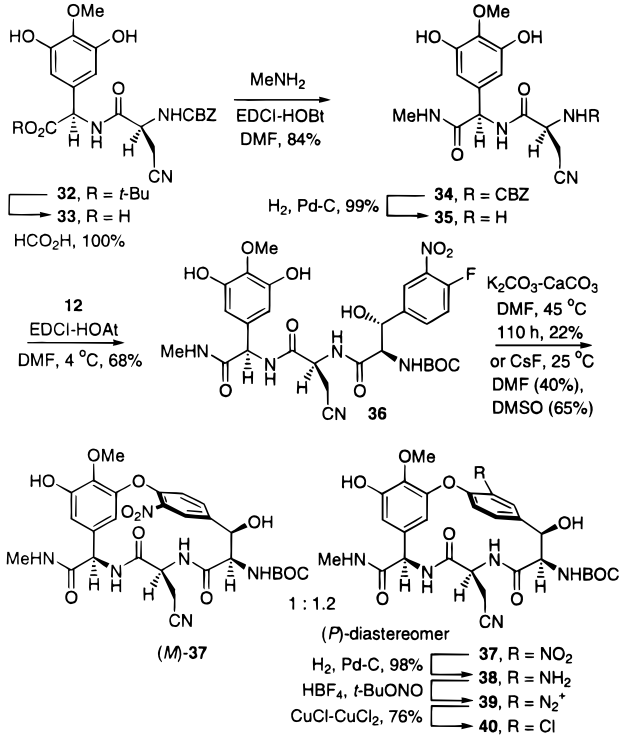
The DE Ring System: A C-Terminus Carboxamide. Independent of these efforts and providing the impetus for the studies described above, the potential for epimerization at the central amino acid phenylglycine C8 site was first implicated in unsuccessful efforts at closing **4**. Prior macrocyclization attempts at the formation of **4** with CsF (5 equiv, 0−25 °C, DMSO, DMF) instead of K_2CO_3/$CaCO_3$ provided the desired products in low conver-

(23) Brown, H. C.; Narasimhan, S. *J. Org. Chem.* **1982**, *47*, 1604.

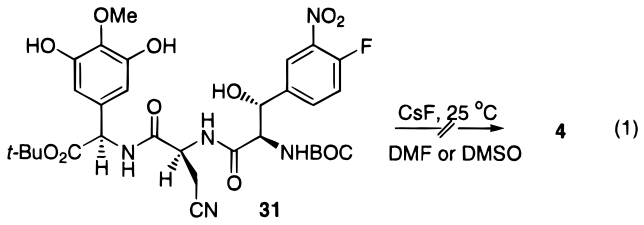
Scheme 5



Scheme 6



sions (ca. 20%) and the major products isolated (70%) implicated additional reactions at the C8 center (eq 1).



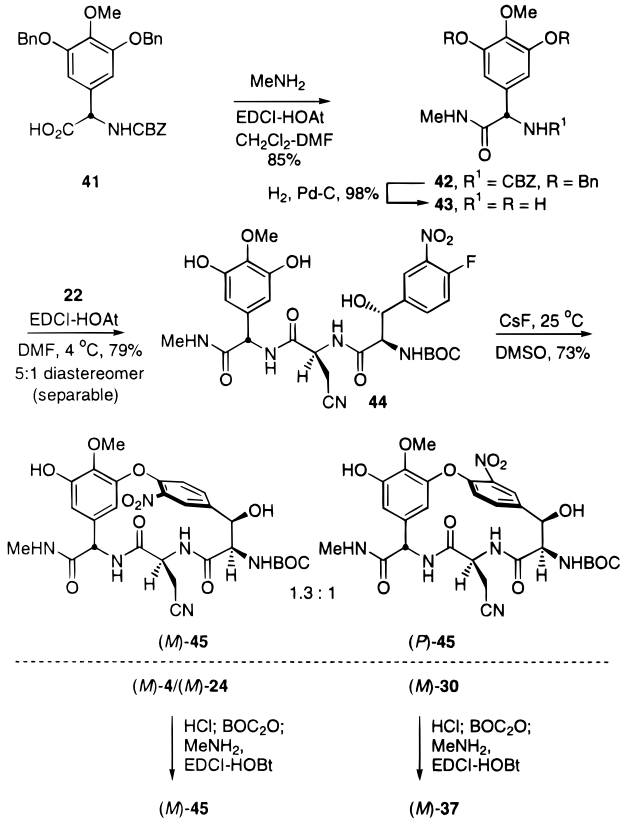
The chemical shifts of C6-H, C21-H, C8-H, and N9-H of the major cyclized products moved by >1 ppm relative to those of **4**, the doublets observed for C8-H and N9-H in **4** collapsed to singlets, and the product atropisomers possessed a molecular weight 16 mass units higher than that of the desired product, suggesting deprotonation and oxidation of the C8 center.[21] These observations contrast those of both Evans and Zhu who have disclosed the use of CsF under mild conditions to promote related macrocyclizations in systems where the C8 center was not substituted with an ester.[8,14]

In efforts that define the source of these distinctions, (8*R*)-*N*-methylcarboxamide **37** and its unnatural (8*S*)-epimer **45** were prepared. The correlation of **45**, not **37**, with **4** served as the first indication that **4** possessed the unnatural 8*S* stereochemistry. Deprotection of the *tert*-butyl ester **32**[11] (HCO_2H, 100%) followed by *N*-methylamide formation (EDCI/HOBt, DMF, 4 °C, 84%) provided **34** as a single detectable diastereomer (Scheme 6). *N*-CBZ deprotection (H_2, Pd/C, 99%), followed by coupling of the free amine **35** with **12** (EDCI/HOAt, DMF, 4 °C, 68%), provided the macrocyclization substrate **36**. Macrocyclization of **36** upon treatment with $K_2CO_3/CaCO_3$ (5.0 equiv/7.5 equiv, 2 wt equiv of 4 Å MS, 0.008 M DMF, 45 °C) was unusually sluggish, providing no product after 24 h, little product after 48 h, and approximately 20% **37** after 110 h. However, CsF-promoted macrocyclization proved successful, affording a 1.2:1 mixture of separable *P* and *M* diastereomers in 65% yield (5.4 equiv CsF, 0.008 M DMSO, 25 °C, 11.5 h), favoring the natural atropisomer. Conducting this reaction in DMF led to substantial amounts of recovered starting material, and the use of DMSO significantly increased the rate of cyclization. In contrast to the closure leading to **4**, the closure of **36** proceeded, without significant C8 epimerization, to provide **37** accompanied by ≤10% of **45**. Reduction of (*P*)-**37** (H_2, Pd/C, CH_3OH, 98%), diazotization (1.3 equiv of HBF_4, 1.3 equiv of *t*-BuONO, CH_3CN, 0 °C, 1−1.5 h), and Sandmeyer substitution (60 equiv of $CuCl_2$, 20 equiv of CuCl, H_2O, 25 °C, 1.5 h, 76%) provided (*P*)-**40** available for final conversion to a fully functionalized vancomycin DE ring system. Notably, the conversions for introduction of the DE aryl chloride have improved considerably over our initial disclosure,[11] as our technical experience with them has matured.

Similarly, coupling of (*S*)-**43**, derived from (*S*)-**41**,[16] with **22**[11] provided **44** (79%), Scheme 7. CsF-promoted macrocyclization (DMSO, 25 °C, 12 h) provided **45** (73%), possessing the unnatural 8*S* stereochemistry, as a separable 1.3:1 mixture of *M*:*P* atropisomers which were readily distinguishable from **37**.

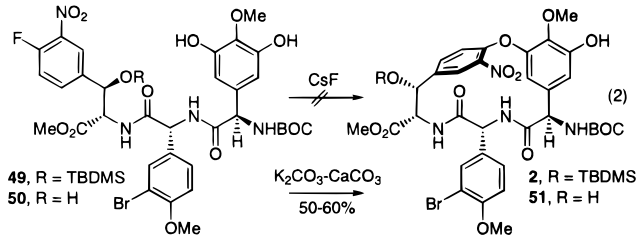
Correlation of **4** and its minor C8 diastereomer **30**[19] with **45** and **37**, respectively, first established the previ-

Scheme 7



ously unrecognized C8 epimerization of **4** (Scheme 7) and led to the initiation of the preceding studies. More importantly, the distinguishable properties of **37** and **45** coupled with their successful correlations with **30**/**4**/**24** and, ultimately, **28**/**29** confirmed that the macrocyclizations of the *N*-methyl carboxamides **36** and **44** proceeded without significant C8 epimerization. Completion of the preparation of the fully functionalized DE ring system involved *N*-BOC deprotection of **40** (3 M HCl/EtOAc, 25 °C, 0.5 h, 100%), coupling the amine hydrochloride **46** with BOCNMe-D-Leu (EDCl/HOAt, 4:1 CH_2Cl_2/DMF, 0 °C, 2 h, 65%), and nitrile hydration of **47** under acidic conditions enlisting moist, saturated HCl/Et_2O,[24] which also served to deprotect the *N*-BOC group providing **48** (Scheme 8).

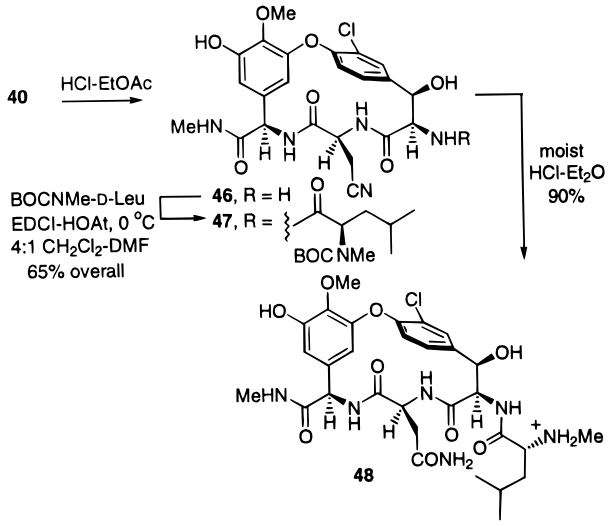
CD Ring System Cyclization. In preceding studies, we also disclosed the macrocyclization of **49**,[11] enlisting K_2CO_3/$CaCO_3$ under conditions where the C15 TBDMS protecting group was not removed (eq 2). Although this



was similarly successful employing the free alcohol, this closure to provide **2** highlighted the effective scavenging of the liberated fluoride by the added $CaCO_3$ and provided

(24) Garcia Ruano, J. L.; Martin Castro, A. M.; Rodriguez, J. H. *J. Org. Chem.* **1992**, *57*, 7235.

Scheme 8

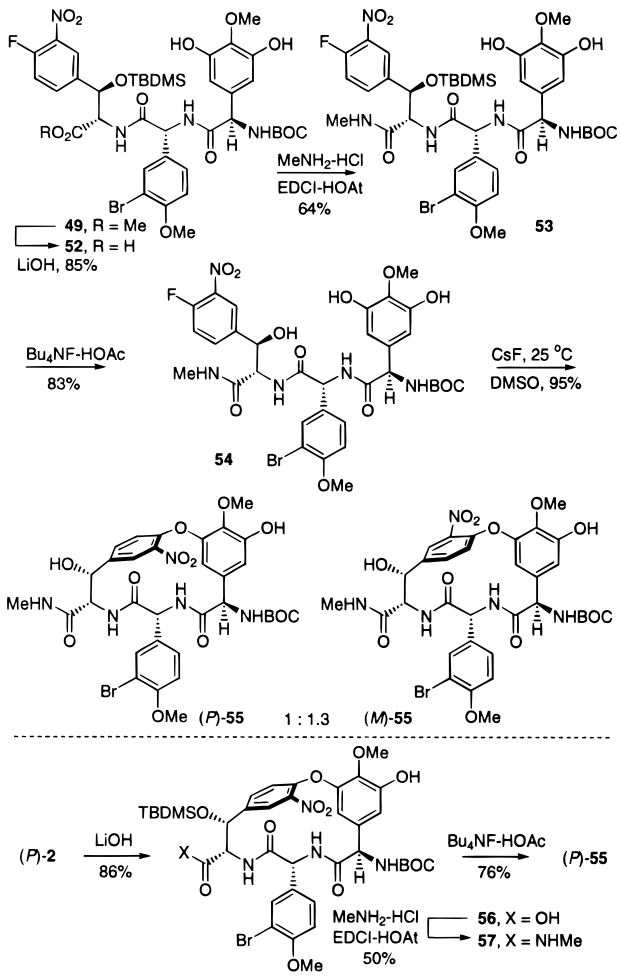


opportunities for thermal atropisomerism of the CD ring system that were not accessible with the labile free alcohol.[10,11] Since the atropisomerism instability of the free alcohol was attributable in part to a thermal retro aldol reaction, a substrate bearing a less acidic C14 center might survive the thermal atropisomerism conditions without such a deliberate alcohol protection.

In addition, efforts to promote the macrocyclization of **49** with CsF (DMF or DMSO, 25 °C, 24 h) failed to provide the desired cyclization products and appeared to provide epimers and/or retro aldol products instead.[11,25] Thus, in conjunction with our thermal atropisomerism studies and as a consequence of the success of the DE macrocyclization with substrates bearing a C8 carboxamide, we examined the macrocyclization substrate **54** substituted with a C14 carboxamide. Low-temperature methyl ester hydrolysis of **49**[11] provided the carboxylic acid **52** (85%) (Scheme 9). Coupling with methylamine (EDCI−HOBt, DMF, 0−5 °C, 24 h, 64%) cleanly afforded **53** with no evidence of epimerization. TBDMS deprotection in the presence of HOAc (6 equiv of Bu_4NF, 5 equiv of HOAc, THF, 0−25 °C, 1.5 h, 83%) provided the cyclization substrate **54** without competitive retro aldol or initiation of the macrocyclization reaction. Closure of **53** or **54** under the K_2CO_3/$CaCO_3$ conditions (5.5 equiv/5.5 equiv, 2 wt equiv of 4 Å MS, DMF, 45 °C) was much slower than the corresponding methyl ester substrate (14 h, 45 °C, 60%),[11] and after 48 h (45 °C), the separable atropisomers of **57** and **55** were isolated in 43% and 32% yield with 22% and 50% of the unreacted starting material recovered, respectively. In the case of the closure of **53**, this occurred without OTBDMS deprotection. In contrast, CsF (5.6 equiv) proved more effective at promoting the cyclization of **54** (0.008 M DMSO, 25 °C, 11

(25) Treatment of **49** with CsF (5.0 equiv, DMSO, 65%, 4:1, 25 °C, 15 h) provided two diastereomeric cyclization products that did not correlate with **51**, and conducting the reaction in DMF provided a second pair of diastereomers (70%, 2.5: 1) that also did not correlate with **51**. For the product derived from the DMF reaction: 1.5: 1 mixture of rotamers: $[\alpha]^{25}_D$ +7 (*c* 0.019, $CHCl_3$); ^{1}H NMR (acetone-d_6, 400 MHz) δ 8.37 (m, 1H), 8.19 (d, 1H, J = 2.1 Hz), 8.03 (m, 1H), 7.90 (dd, 1H, J = 8.4, 1.9 Hz), 7.01 (m, 1H), 7.41 (dd, 1H, J = 8.4, 2.0 Hz), 7.12 (d, 1H, J = 8.5 Hz), 6.69 (m, 1H), 5.74−5.67 (m, 2H), 5.53−5.37 (m, 1H), 3.96 (s, 3H), 3.86 (s, 3H), 3.73 (s, 3H), 1.22 (s, 9H), 0.44 (s, 9H); IR (film) ν_{max} 3410, 2956, 1742, 1710, 1680, 1580, 1530, 1495, 1343, 1255, 1100 cm^{-1}; FABHRMS (NBA) *m*/*z* 907.0471 (M^+ + Cs, $C_{33}H_{35}O_{13}N_{14}Br$ requires 907.0438).

Scheme 9



h, 95%; 0.008 M DMF, 25 °C, 34 h, 53%), providing a 1:1.3 mixture of separable atropisomers with the *M* isomer predominating slightly. Similarly, subjecting **53** to CsF promoted macrocyclization (8 equiv, DMSO, 25 °C, 15 h) and also provided the same 1:1.3 mixture of **55** atropisomers (64%) resulting from both ring closure and TBDMS deprotection. The atropisomer stereochemistry and confirmation of the maintained C15/C14 stereochemistry was established by 2D $^1H-^1H$ NMR of (*P*)-**55**, which exhibited diagnostic H-17/H-15 (s) and H-17/H-14 (s) NOE's as is characteristic of the natural atropisomer stereochemistry and lacked H-20/H-15, H-20/H-14 NOE's as is diagnostic of the unnatural atropisomer. The integrity of the remaining stereocenters was established by conversion of authentic (*P*)-**2** to (*P*)-**55** (Scheme 9).[26] This provided independent confirmation of the stereochemical integrity of **2** and the improved C14 carboxamide cyclization (CsF, DMSO, 95%) provides second-generation improvements in the synthesis of the vancomycin CD ring system, applicable to the natural product itself. Consistent with prior observations, the CD ring closure typically is better than the DE ring closure, and the relative effectiveness of the **36** and **54** macrocyclizations reflect this trend.

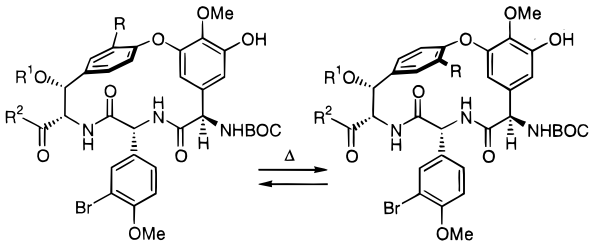
Thermal Atropisomerism. We have disclosed that the thermal interconversion of the CD and DE ring system atropisomers proceeds rapidly at temperatures of ≥155 °C and more slowly at 140 °C. This allows the undesired atropisomers to be thermally equilibrated, chromatographically reisolated, and recycled to provide the desired atropisomers. The precursor CD and DE nitro derivatives were found to equilibrate more rapidly than the corresponding chloro atropisomers, and the rate of isomerization could be controlled not only by the choice of temperature but also by the choice of solvent. Significantly, the equilibration of the DE atropisomer occurred much more rapidly than that of the CD ring system. This suggested that it would be possible to equilibrate the DE, as opposed to the CD, atropisomers within an intact CDE ring system or the ABCDE ring system of vancomycin itself and that this would be best conducted with a DE aryl nitro intermediate containing the installed CD aryl chloride.[11] This has been successfully implemented[10,27] and suggested a preferred order to the synthetic introduction of the CD and DE ring systems in which the CD ring system is assembled first and the atropisomer stereochemistry set with equilibration of its aryl nitro derivative. Following conversion to the CD aryl chloride, introduction of the DE ring system and subsequent selective DE atropisomer equilibration provides a unique solution to the control of vancomycin CDE atropisomer stereochemistry.[9] Consequently, the thermal atropisomerism of the substituted CD and DE ring systems disclosed herein was examined to further define its scope.

The thermal equilibration of **55** and **57** was examined for comparison with **2**, **3**, and **51**. In preceding studies,[10,11] the protected alcohols **2** and **3** were found to most rapidly equilibrate in DMSO and DMF and to do so much more slowly in *o*-$Cl_2C_6H_4$ (Table 1). Even in DMSO, little or no equilibration was observed at 120 °C, slow equilibration was observed at 140 °C, and rapid equilibration was observed at 155 °C. The nitro derivative **2** equilibrated more rapidly than the corresponding chloro derivative **3**. In contrast, the free alcohol **51**[11] failed to undergo clean atropisomerism in either DMSO or *o*-$Cl_2C_6H_4$ (140 °C).[10] Although this was not investigated in detail, retro aldol ring cleavage contributed in part to this instability. The retro aldol product (>10%) and five additional minor products appear early in the thermal isomerism, and comparable side reactions are not observed with **2** or **3**. This led to initial adoption of a synthetic approach to the CD and CDE ring systems that incorporates and preserves protection of the C15 alcohol despite challenges this introduces for macrocyclization via biaryl ether formation.[9,11,27] One important element of thermal atropisomerism that we wanted to examine with **55** was the potential impact that the lower acidity of the C14 position, resulting from the carboxamide substitution, would have on the competitive retro aldol reaction of the free alcohol. Like **51**, thermal equilibration attempts with **55** in DMSO (140 °C) provided only retro aldol products, and no atropisomer was detected by ^1H NMR. In contrast to **51**, **55** was found to cleanly thermally equilibrate in *o*-$Cl_2C_6H_4$ without competitive retro aldol, to do so slightly more rapidly than the corresponding TBDMS-protected alcohol, and to provide a slightly more favorable ratio of atropisomers. All three of these attributes of the thermal atropisomerism of **55**, along with its slow rate relative

(26) The stereochemical integrity of the C14 center was independently established by preparation and cyclization of the C14 hydroxymethyl derivative corresponding to **50** and the correlation of the product atropisomers with **51**. Details are provided in supporting information.

(27) Boger, D. L.; Miyazaki, S.; Loiseleur, O.; Beresis, R. T.; Castle, S. L.; Wu, J. H.; Jin, Q. *J. Am. Chem. Soc.* **1998**, *120*, 8920.

Table 1



2, R = NO_2 R^1 = TBDMS R^2 = OMe 1.1 : 1
3, R = Cl R^1 = TBDMS R^2 = OMe 1.2 : 1
51, R = NO_2 R^1 = H R^2 = OMe 1.6 : 1[a]
57, R = NO_2 R^1 = TBDMS R^2 = NHMe 1.1 : 1
55, R = NO_2 R^1 = H R^2 = NHMe 1.05 : 1

for **2** (DMSO): E_a = 26.6 kcal/mol, $\Delta H^‡$= 27.0 kcal/mol, $\Delta S^‡$ = -1.7 eu, $\Delta G^‡$ (130 °C) = 27.7 kcal/mol

for **3** (*o*-$Cl_2C_6H_4$): E_a = 30.4 kcal/mol, $\Delta H^‡$ = 29.7 kcal/mol, $\Delta S^‡$ = -2.7 eu, $\Delta G^‡$ (140 °C) = 30.8 kcal/mol

for **55** (*o*-$Cl_2C_6H_4$): E_a = 26.9 kcal/mol, $\Delta H^‡$ = 25.9 kcal/mol, $\Delta S^‡$ = -2.6 eu, $\Delta G^‡$ (140 °C) = 27.0 kcal/mol

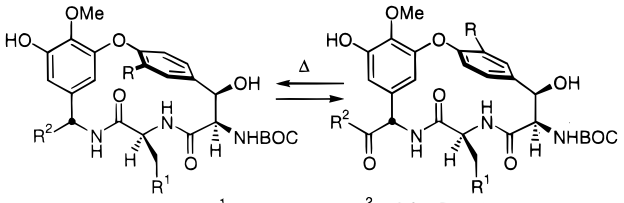
compd	conditions	k (h^{-1})	$t_{1/2}$ (h)
2	155 °C, DMSO	0.27	1.06
2	140 °C, DMSO	0.082	3.52
2	140 °C, *o*-$Cl_2C_6H_4$	0.029	9.77
3	140 °C, DMSO	0.071	4.03
3	140 °C, *o*-$Cl_2C_6H_4$	0.0065	44.0
3	150 °C, *o*-$Cl_2C_6H_4$	0.0156	14.2
51	140 °C, DMSO	0.086	2.52[a]
51	140 °C, *o*-$Cl_2C_6H_4$	*b*	
55	135 °C, *o*-$Cl_2C_6H_4$	0.034	8.34
55	140 °C, *o*-$Cl_2C_6H_4$	0.041	6.56
55	140 °C, DMSO	*c*	
57	140 °C, *o*-$Cl_2C_6H_4$	0.031	9.22

[a] Accompanied by decomposition, data approximate. [b] Extensive decomposition. [c] Only retro aldol observed.

to the DE ring system and the improved macrocyclization (95%), define significant improvements for second-generation syntheses of the CDE ring system and of the natural product itself.

The thermal atropisomer equilibrations of the new series of DE ring system derivatives as well as the previously disclosed derivatives[10,11] are summarized in Table 2. Among these are the authentic 8*R* derivatives, the initial 8*S* derivatives incorporating the Asn residue carboxamide protected as a nitrile, as well as the derivatives disclosed herein incorporating the free and Ddm-protected carboxamide. Unlike the CD ring system, all the derivatives which contain the free C15 alcohol were found to withstand thermal atropisomerism conditions without evidence of competitive retro aldol ring cleavage. Analogous to observations made with the CD ring system, the nitro derivatives equilibrate more readily than the corresponding chloro derivatives. Unlike the CD ring system, the equilibration was remarkably rapid and proceeded readily even at 110−130 °C, and the unnatural 8*S* derivatives isomerize more rapidly than the natural 8*R* derivatives. Because of the rapid rates, the choice of solvent has a less pronounced effect on the apparent ease of atropisomerism. The nature of the Asn-residue substituent had little impact on this rapid rate of atropisomerism, and the Ddm-protected carboxamide **21** as well as the free carboxamide **58**[28] isomerized at rates comparable with **4**. In fact, the rate of isomerization of the free carboxamide **58** was slightly faster than that of the corresponding nitrile ($t_{1/2}$ 5.5 min, 140 °C, DMSO). This

Table 2



4, R = NO_2 R^1 = CN R^2 = CO_2t-Bu 1 : 1
5, R = Cl R^1 = CN R^2 = CO_2t-Bu 1 : 1
21, R = NO_2 R^1 = CONHDdm R^2 = CO_2t-Bu 1.3 : 1
29, R = NO_2 R^1 = CN R^2 = CH_2OH 1.1 : 1
58, R = NO_2 R^1 = $CONH_2$ R^2 = CO_2t-Bu 1.3 : 1[27]
45, R = NO_2 R^1 = CN R^2 = CONHMe 1 : 1

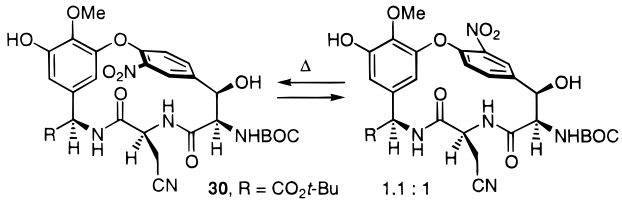
for **4** (DMSO): E_a = 15.3 kcal/mol, $\Delta H^‡$ = 14.5 kcal/mol $\Delta S^‡$ = -24.0 eu, $\Delta G^‡$(130 °C) = 24.2 kcal/mol

for **4** (*o*-$Cl_2C_6H_4$): E_a = 16.4 kcal/mol, $\Delta H^‡$ = 15.2 kcal/mol $\Delta S^‡$ = -22.0 eu, $\Delta G^‡$(130 °C) = 24.1 kcal/mol

for **21** (DMSO): E_a = 17.1 kcal/mol, $\Delta H^‡$ = 16.8 kcal/mol $\Delta S^‡$ = -17.3 eu, $\Delta G^‡$(130 °C) = 23.8 kcal/mol

for **29** (DMSO): E_a = 16.7 kcal/mol, $\Delta H^‡$ = 15.9 kcal/mol $\Delta S^‡$ = -20.0 eu, $\Delta G^‡$(130 °C) = 24.0 kcal/mol

for **58** (DMSO): E_a = 18.5 kcal/mol, $\Delta H^‡$ = 17.9 kcal/mol $\Delta S^‡$ = -14.3 eu, $\Delta G^‡$(130 °C) = 23.7 kcal/mol



30, R = CO_2t-Bu 1.1 : 1
28, R = CH_2OH 1 : 1
37, R = CONHMe 1 : 1

for **30** (DMSO): E_a = 21.1 kcal/mol, $\Delta H^‡$ = 20.6 kcal/mol $\Delta S^‡$ = -11.6 eu, $\Delta G^‡$(130 °C) = 25.2 kcal/mol

for **28** (DMSO): E_a = 18.8 kcal/mol, $\Delta H^‡$ = 18.3 kcal/mol $\Delta S^‡$ = -16.3 eu, $\Delta G^‡$(130 °C) = 24.9 kcal/mol

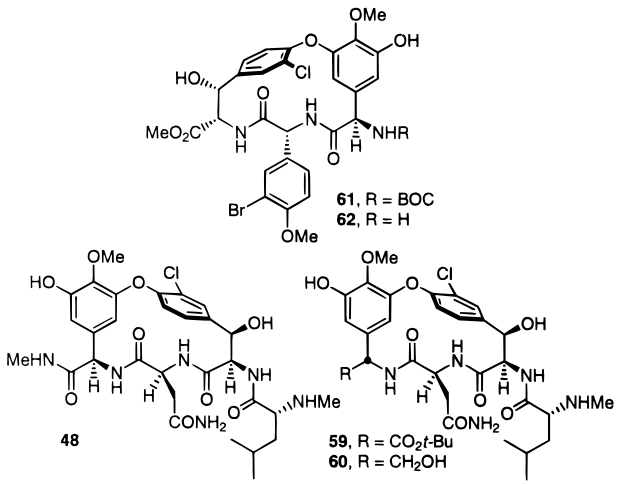
for **37** (DMSO): E_a = 23.6 kcal/mol, $\Delta H^‡$ = 22.9 kcal/mol $\Delta S^‡$ = -6.1 eu, $\Delta G^‡$(130 °C) = 25.4 kcal/mol

compd	conditions	k (h^{-1})	$t_{1/2}$ (h)
4	130 °C, DMSO	0.66	0.23
4	140 °C, DMSO	1.05	0.17
4	130 °C, *o*-$Cl_2C_6H_4$	0.70	0.29
4	140 °C, *o*-$Cl_2C_6H_4$	1.15	0.15
5	140 °C, DMSO	0.52	0.39
21	120 °C, DMSO	0.58	0.43
21	130 °C, DMSO	1.5	0.17
21	140 °C, DMSO	1.7	0.15
29	130 °C, DMSO	0.90	0.29
29	140 °C, DMSO	1.5	0.16
58	125 °C, DMSO	0.88	0.28
58	140 °C, DMSO	2.06	0.092
45	140 °C, DMSO	1.28	0.11
30	130 °C, DMSO	0.17	1.6
30	140 °C, DMSO	0.33	0.83
28	130 °C, DMSO	0.28	1.0
28	140 °C, DMSO	0.49	0.59
37	130 °C, DMSO	0.16	1.9
37	140 °C, DMSO	0.32	1.0

thermal atropisomerism of the free carboxamide **58** was observed without competitive Asn−isoaspartate rearrangement analogous to that observed in the acidic,

(28) The amide **58** was prepared by treatment of **24** with H_2O_2/K_2CO_3 (4 equiv/2.3 equiv, 2:1 DMSO/H_2O, 45 °C, 1.5 h, 64%) without competitive ester hydrolysis. Katritzky, A. R.; Pilarski, B.; Urogdi, L. *Synthesis* **1989**, 949.

Table 3. Antimicrobial Activity, MIC (mg/mL)



compd	*Staphylococcus aureus*[a]	*Enterococcus faecium*[b]	*Enterococcus faecium*[c]
1	1.25	2.5	250
48	>1050	>1050	>1050
59	1250	>2500	>2500
60	>940	>940	>940
61	>1500	>1500	>1500
62	>2100	>2100	>2100

[a] ATCC 25923. [b] ATCC 35667. [c] Vancomycin resistant.

thermal degradation (pH 4.2, 70−80 °C, 40 h) studies of vancomycin leading to CDP-1[2] and, in part, may be attributed to a rapid isomerism precluding competitive rearrangement.[27] The observation that the natural 8*R* derivatives isomerize more slowly than the 8*S* derivatives is consistent with our disclosure that DE atropisomerism within both the intact CDE ring system[9] and within vancomycin[27] proceeds slowly and at a rate ca. 3× slower than in **37** itself. This may now be attributed in part to the (8*R*)-carboxamide stereochemistry present in the CDE ring systems.

Antimicrobial Activity. Three fully functionalized DE ring systems incorporating the aryl chloride, the natural atropisomer stereochemistry, the Asn free carboxamide, and the MeNH-D-Leu side chain including the analogues bearing the natural C8 *N*-methylcarboxamide and the unnatural C8 hydroxmethyl and *tert*-butyl ester[29] substituents as well as two fully functionalized CD ring systems were tested for antimicrobial activity in vancomycin-sensitive and -resistant bacteria. In all cases, they were ≥1000× less potent than vancomycin, and only **59** exhibited a detectable activity at this level (Table 3).

Experimental Section

***N*-Methyl (*P*)- and (*M*)-(8*R*,11*S*,14*R*,15*R*)-14-[*N*-[(*tert*-Butyloxy)carbonyl]amino]-11-(cyanomethyl)-5,15-dihydroxy-10,13-dioxo-4-methoxy-18-nitro-9,12-diaza-2-oxatricyclo[14.2.2.1^{3,7}]heneicosa-3,4,7-(21),-16,18,19-hexaene-8-carboxamide (37).** A solution of **36** (9.4 mg, 0.014 mmol) in anhydrous degassed DMSO (1.8 mL) was added to a vial charged with predried (130 °C, 0.1 mm Hg, 12 h) CsF (12.0 mg, 0.079 mmol, 5.4 equiv). The mixture was stirred at 25 °C for 11.5 h. The resulting mixture was filtered through a pad of Celite (10% $CH_3OH/CHCl_3$) and concentrated in vacuo. PTLC (SiO_2, 10% $CH_3OH/CHCl_3$, three elutions) afforded **37** as a separable 1.2:1 mixture of diastereomers (5.9 mg, 9.1 mg theoretical, 65%). For the major, more polar *P* diastereomer: white film (3.2 mg, 35%); $[\alpha]^{25}_D$ +8.3 (*c* 0.12, CH_3OH); ^{1}H NMR (CD_3OD, 400 MHz) δ 8.15 (s, 1H), 7.81 (dd, 1H, J = 2.0, 8.5 Hz), 7.18 (d, 1H, J = 8.5 Hz), 6.57−6.55 (m, 1H), 5.60−5.58 (m, 1H), 5.15 (d, 1H, J = 3.2 Hz), 4.89−4.86 (m, 1H, partially obscured by H_2O), 4.60 (m, 1H), 4.32 (d, 1H, J = 3.3 Hz), 3.96 (s, 3H), 2.80 (dd, 1H, J = 6.7, 16.7 Hz), 2.73−2.67 (m, 1H), 2.69 (s, 3H), 1.48 (s, 9H); ^{13}C NMR (CD_3OD, 100 MHz) δ 171.5, 171.2, 169.2, 154.7, 152.5, 150.0, 144.2, 140.8, 138.0, 133.3, 133.1, 129.5, 127.3, 124.6, 117.5, 110.9, 106.0, 81.2, 73.9, 61.7, 60.6, 59.0, 51.1, 28.7 (3C), 26.7, 21.7; IR (film) ν_{max} 3289, 2930, 1653, 1534, 1349, 1239, 1164, 1086, 1027 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 761.1157 (M^+ + Cs, $C_{28}H_{32}N_6O_{11}$ requires 761.1183). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 40 °C) of (*P*)-**37** displayed the following diagnostic NOE cross-peaks: C17-H/C15-H (s, δ 8.15/5.15), C17-H/C14-H (s, δ 8.15/4.32), C20-H/C19-H (m, δ 7.81/7.18), C6-H/C8-H (m, δ 6.57−6.55/4.89−4.86), C15-H/C14-H (m, δ 5.15/4.32), C11-H/CH_2CN (w, δ 4.60/2.80 and 4.60/2.73−2.67).

For the minor, less polar *M* diastereomer: white film (2.7 mg, 30%); $[\alpha]^{25}_D$ +35 (*c* 0.14, CH_3OH); ^{1}H NMR (CD_3OD, 400 MHz) δ 8.16 (s, 1H), 7.80−7.77 (m, 1H), 7.38 (d, 1H, J = 8.3 Hz), 6.51−6.49 (m, 1H), 5.46−5.42 (m, 1H), 5.16 (d, 1H, J = 3.7 Hz), 4.90−4.87 (m, 1H, partially obscured by H_2O), 4.61−4.56 (m, 1H), 4.45 (d, 1H, J = 3.7 Hz), 3.96 (s, 3H), 2.83−2.78 (m, 1H), 2.76−2.71 (m, 1H), 2.70 (s, 3H), 1.48 (s, 9H); IR (film) ν_{max} 3290, 2921, 2849, 1652, 1539, 1507, 1153 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 761.1158 (M^+ + Cs, $C_{28}H_{32}N_6O_{11}$ requires 761.1183). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 40 °C) of (*M*)-**37** displayed the following diagnostic NOE cross-peaks: C20-H/C19-H (s, δ 7.80−7.77/7.38), C20-H/C15-H (s, δ7.80−7.77/5.16), C20-H/C14-H (s, δ 7.80−7.77/4.45), C6-H/C8-H (m, δ 4.90−4.87/6.51−6.49), C11-H/CH_2CN (w, δ 4.61−4.56/2.83−2.78 and 4.61−4.56/2.76−2.71).

***N*-Methyl (*P*)-(8*R*,11*S*,14*R*,15*R*)-18-Amino-14-[*N*-[(*tert*-butyloxy)carbonyl]amino]-11-(cyanomethyl)-5,15-dihydroxy-10,13-dioxo-4-methoxy-9,12-diaza-2-oxatricyclo[14.2.2.1^{3,7}]heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxamide (38).** A solution of (*P*)-**37** (3.0 mg, 0.0048 mmol) in anhydrous CH_3OH (0.3 mL) was treated with 10% Pd/C (0.4 mg, 0.13 wt equiv) and stirred at 25 °C under H_2 (1 atm) for 4 h. The reaction mixture was filtered through a pad of Celite (washed with 10% $CH_3OH/CHCl_3$) and concentrated in vacuo to afford **38** (2.8 mg, 2.9 mg theoretical, 98%) as a white film: ^{1}H NMR (CD_3OD, 400 MHz) δ 6.85−6.83 (m, 1H), 6.80−6.78 (m, 1H), 6.76 (s, 1H), 6.50 (d, 1H, J = 2.1 Hz), 5.70−5.67 (m, 1H), 5.02 (s, 1H), 4.93 (d, 1H, J = 5.6 Hz), 4.65−4.60 (m, 1H), 4.25 (d, 1H, J = 3.4 Hz), 3.97 (s, 3H), 2.81 (dd, 1H, J = 6.4, 16.9 Hz), 2.72 (dd, 1H, J = 6.2, 16.9 Hz), 2.70 (s, 3H), 1.48 (s, 9H); FABHRMS (NBA-CsI) *m*/*z* 731.1462 (M^+ + Cs, $C_{28}H_{34}N_6O_9$ requires 731.1442).

***N*-Methyl (*P*)-(8*R*,11*S*,14*R*,15*R*)-14-[*N*-[(*tert*-Butyloxy)carbonyl]amino]-18-chloro-11-(cyanomethyl)-5,15-dihydroxy-10,13-dioxo-4-methoxy-9,12-diaza-2-oxatricyclo[14.2.2.1^{3,7}]heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxamide (40).** A solution of **38** (4.2 mg, 0.0070 mmol) in anhydrous CH_3CN (0.25 mL) at 0 °C was treated with HBF_4 (0.1 M solution in CH_3CN, 91 μL,

(29) Details of the preparation of **59** and **60** may be found in the Supporting Information.

0.0091 mmol, 1.3 equiv), and the resulting solution was stirred at 0 °C for 10 min before being warmed to 25 °C and stirred for 30 min. The reaction mixture was recooled to 0 °C, treated with *tert*-butyl nitrite (0.1 M solution in CH_3CN, 91 μL, 0.0091 mmol, 1.3 equiv), and stirred at 0 °C for 1 h. The solvent was removed in vacuo at 0 °C, and the residue containing **39** was treated with a solution of $CuCl_2$ (57 mg, 0.42 mmol, 60 equiv) in H_2O (0.55 mL). The resulting mixture was stirred at 25 °C for 5 min, treated with CuCl (14 mg, 0.14 mmol, 20 equiv), and stirred at 25 °C for 1.5 h. Saturated aqueous NH_4Cl (1.5 mL) was added to the mixture, and it was extracted with EtOAc (5 × 1.5 mL). The combined organic extracts were washed with H_2O (2.5 mL), saturated aqueous NH_4Cl (2.5 mL), and saturated aqueous NaCl (2.5 mL), dried (Na_2SO_4), and concentrated in vacuo. PTLC (SiO_2, eluted twice with 10% $CH_3OH/CHCl_3$) afforded **40** (3.3 mg, 4.3 mg theoretical, 76%) as a white film: $[\alpha]^{25}_D$ +23 (*c* 0.1, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 7.60 (s, 1H), 7.47 (dd, 1H, J = 1.9, 8.4 Hz), 7.07 (d, 1H, J = 8.5 Hz), 6.53 (d, 1H, J = 1.8 Hz), 5.49−5.46 (m, 1H), 5.05 (d, 1H, J = 2.8 Hz), 4.96 (s, 1H), 4.62−4.58 (m, 1H), 4.28 (d, 1H, J = 3.5 Hz), 3.97 (s, 3H), 2.79 (dd, 1H, J = 6.8, 16.4 Hz), 2.73−2.67 (m, 1H), 2.71 (s, 3H), 1.48 (s, 9H); IR (film) ν_{max} 3302, 2919, 1658, 1512, 1237, 1161, 1038 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 750.0919 (M^+ + Cs, $C_{28}H_{32}N_5O_9Cl$ requires 750.0943).

***N*-Methyl (*P*)-(8*R*,11*S*,14*R*,15*R*)-14-Amino-18-chloro-11-(cyanomethyl)-5,15-dihydroxy-10,13-dioxo-4-methoxy-9,12-diaza-2-oxatricyclo[14.2.2.1^{3,7}]heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxamide (46).** A vial charged with **40** (3.3 mg, 0.0053 mmol) was treated with 3 M HCl/EtOAc (0.5 mL), and the resulting mixture was stirred at 25 °C for 30 min. The mixture was concentrated in vacuo to afford **46** (2.8 mg, 2.8 mg theoretical, quantitative) as a white film which was used directly in the next reaction: ^1H NMR (CD_3OD, 400 MHz) δ 7.56 (d, 1H, J = 1.9 Hz), 7.49 (dd, 1H, J = 2.1, 8.3 Hz), 7.11 (d, 1H, J = 8.4 Hz), 6.51 (d, 1H, J = 2.2 Hz), 5.64 (d, 1H, J = 2.1 Hz), 5.19 (d, 1H, J = 4.3 Hz), 4.78 (s, 1H), 4.46−4.43 (m, 1H), 4.16 (d, 1H, J = 4.3 Hz), 3.98 (s, 3H), 2.91 (dd, 1H, J = 6.5, 17.1 Hz), 2.82 (dd, 1H, J = 4.6, 17.1 Hz), 2.70 (s, 3H).

***N*-Methyl (*P*)-(8*R*,11*S*,14*R*,15*R*)-14-[*N*-[*N*-[(*tert*-Butyloxy)carbonyl]-*N*-methyl]-D-leucyl]amino-18-chloro-11-(cyanomethyl)-5,15-dihydroxy-10,13-dioxo-4-methoxy-9,12-diaza-2-oxatricyclo[14.2.2.1^{3,7}]-heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxamide (47).** A solution of **46** (2.6 mg, 0.0050 mmol) in CH_2Cl_2/DMF (4:1, 0.38 mL) at 0 °C was treated with BOC-NMe-D-Leu (1.8 mg, 0.0073 mmol, 1.5 equiv) and $NaHCO_3$ (0.5 mg, 0.006 mmol, 1.2 equiv) and stirred at 0 °C for 10 min. The mixture was then treated with HOAt (1.5 mg, 0.011 mmol, 2.2 equiv) and EDCI·HCl (1.9 mg, 0.0099 mmol, 2.0 equiv) and stirred at 0 °C under Ar for 2 h. The mixture was treated with H_2O (1 mL) and extracted with EtOAc (5 × 1.5 mL). The combined organic extracts were washed with H_2O (2 mL) and saturated aqueous NaCl (2 mL), dried (Na_2SO_4), and concentrated in vacuo. PTLC (SiO_2, eluted twice with 10% $CH_3OH/CHCl_3$) afforded **47** (2.4 mg, 3.7 mg theoretical, 65%) as a white film: $[\alpha]^{25}_D$ +46 (*c* 0.095, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 7.60 (d, 1H, J = 1.8 Hz), 7.51−7.47 (m, 1H), 7.08 (d, 1H, J = 8.4 Hz), 6.52 (d, 1H, J = 1.8 Hz), 5.49 (d, 1H, J = 1.6 Hz), 5.06 (d, 1H, J = 3.3 Hz), 4.93 (s, 1H), 4.90−4.87 (m, 1H, partially obscured by H_2O), 4.62−4.57 (m, 2H), 3.97 (s, 3H), 2.81−2.74 (m, 1H, partially obscured), 2.79 (s, 3H), 2.72−2.67 (m, 1H, partially obscured), 2.71 (s, 3H), 1.75−1.69 (m, 2H), 1.51−1.44 (m, 1H, partially obscured), 1.51 (s, 9H), 1.00−0.93 (m, 6H); IR (film) ν_{max} 3820, 3743, 3674, 1771, 1717, 1699, 1652, 1558, 1539, 1506 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 877.1970 (M^+ + Cs, $C_{35}H_{45}N_6O_{10}Cl$ requires 877.1940).

***N*-Methyl (*P*)-(8*R*,11*S*,14*R*,15*R*)-14-[*N*-(*N*-Methyl)-D-leucyl]amino-18-chloro-11-ethanamido-5,15-dihydroxy-10,13-dioxo-4-methoxy-9,12-diaza-2-oxatricyclo-[14.2.2.1^{3,7}]heneicosa-3,4,7(21),16,18,19-hexaene-8-carboxamide (48).** A vial charged with **47** (1.1 mg, 0.0015 mmol) at 0 °C was treated with a saturated solution of HCl in Et_2O (0.4 mL) followed by H_2O (10 μL), and the mixture was stirred at 25 °C for 73 h. The mixture was cooled to 0 °C, concentrated under a stream of N_2, and purified by reverse-phase HPLC (C18, CH_3CN−0.07% aqueous TFA 23:77 as eluant) to afford **48** (0.9 mg, 1.0 mg theoretical, 90%) as a white film: $[\alpha]^{25}_D$ +16 (*c* 0.070, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 7.60−7.57 (m, 1H), 7.53−7.49 (m, 1H), 7.11 (d, 1H, J = 8.5 Hz), 6.55 (d, 1H, J = 1.8 Hz), 5.46 (d, 1H, J = 1.8 Hz), 5.08−5.05 (m, 1H), 5.00 (s, 1H), 4.90−4.87 (m, 1H, obscured by H_2O), 4.68−4.63 (m, 1H), 4.57 (d, 1H, J = 3.6 Hz), 3.97 (s, 3H), 2.71 (s, 3H), 2.67 (s, 3H), 2.58−2.53 (m, 1H), 2.50−2.44 (m, 1H), 1.79−1.64 (m, 2H), 1.38−1.31 (m, 1H), 1.07 (d, 3H, J = 6.4 Hz), 1.03 (d, 3H, J = 6.4 Hz); IR (film) ν_{max} 3319, 1673, 1650, 1434, 1205, 1138 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 795.1542 (M^+ + Cs, $C_{30}H_{39}N_6O_9Cl$ requires 795.1521).

(2*S*,3*R*)-3-[(*tert*-Butyldimethylsilyl)oxy]-2-[*N*-[(*R*)-*N*-[(*R*)-*N*-[(*tert*-butyloxy)carbonyl]-(3,5-dihydroxy-4-methoxyphenyl)glycyl]-(3-bromo-4-methoxyphenyl)-glycyl]amino]-3-(4-fluoro-3-nitrophenyl)-propionic Acid (52). A solution of **49**[11] (180 mg, 0.198 mmol) in *t*-BuOH/H_2O (2:1, 6.0 mL) was treated with $LiOH\cdot H_2O$ (16.6 mg, 0.40 mmol, 2.0 equiv) at 0 °C under Ar for 3 h. The reaction mixture was quenched with the addition of 15% aqueous citric acid (5 mL) at 0 °C and the volatiles were removed in vacuo. The aqueous phase was extracted with EtOAc (3 × 10 mL). The combined organic phases were washed with H_2O (10 mL) and saturated aqueous NaCl (10 mL), dried (Na_2SO_4), and concentrated in vacuo. PTLC (SiO_2, 10% $CH_3OH/CHCl_3$) afforded **52** (150 mg, 85%) as a pale yellow solid: $[\alpha]^{25}_D$ −31 (*c* 0.71, CH_3OH); ^1H NMR (acetone-d_6, 400 MHz) δ 8.01 (s, 2H), 7.77 (d, 1H, J = 8.1 Hz), 7.59 (d, 1H, J = 8.1 Hz), 7.48 (br s, 2H), 7.30−7.22 (m, 2H), 7.18−6.81 (m, 2H), 6.59 (s, 1H), 6.55 (s, 1H), 6.36 (s, 1H), 5.60 (s, 1H), 5.45 (s, 1H), 5.19 (br s, 1H), 4.58 (d, 1H, J = 9.0 Hz), 3.90 (s, 3H), 3.76 (s, 3H), 1.36 (s, 9H), 0.77 (s, 9H), −0.06 (s, 3H), −0.20 (s, 3H); IR (neat) ν_{max} 3292, 2947, 1687, 1678, 1603, 1537, 1497, 1366, 1259, 1165 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 1027.1463 (M^+ + Cs, $C_{38}H_{48}N_4O_{13}BrFSi$ requires 1027.1454).

***N*-Methyl (2*S*,3*R*)-3-[(*tert*-Butyldimethylsilyl)-oxy]-2-[*N*-[(*R*)-*N*-[(*R*)-*N*-[(*tert*-butyloxy)carbonyl]-(3,5-dihydroxy-4-methoxyphenyl)glycyl]-(3-bromo-4-methoxyphenyl)glycyl]amino]-3-(4-fluoro-3-nitrophenyl)propionamide (53).** A solution of **52** (53 mg, 0.060 mmol) in anhydrous DMF (2.0 mL) was treated sequentially with HOBt (20.5 mg, 0.15 mmol, 2.5 equiv), $CH_3NH_2\cdot HCl$ (20 mg, 0.30 mmol, 5.0 equiv), $NaHCO_3$ (25 mg, 0.30 mmol, 5.0 equiv), and EDCI (28 mg, 0.144 mmol, 2.4 equiv) at 0 °C under Ar and stirred for 24 h. After removal of the solvent in vacuo, H_2O (5 mL) and EtOAc

(5 mL) were added to the residue. The aqueous phase was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with H_2O (10 mL) and saturated aqueous NaCl (10 mL), dried (Na_2SO_4), and concentrated in vacuo. PTLC (SiO_2, 5% $CH_3OH/CHCl_3$) afforded **53** (35 mg, 65%) as a yellow solid: $[\alpha]^{25}_D$ −16 (*c* 0.60, $CHCl_3$); ^{1}H NMR (acetone-d_6, 400 MHz) δ 8.07 (d, 1H, J = 7.2 Hz), 8.02 (s, 2H), 7.95 (s, 1H), 7.67 (d, 1H, J = 9.1 Hz), 7.63 (br s, 1H), 7.52 (m, 1H), 7.47 (d, 1H, J = 2.1 Hz), 7.18−7.12 (m, 2H), 6.90 (d, 1H, J = 8.4 Hz), 6.46 (s, 2H), 6.30 (d, 1H, J = 7.1 Hz), 5.57 (s, 1H), 5.44 (d, 1H, J = 6.4 Hz), 5.15 (d, 1H, J = 6.0 Hz), 4.54 (dd, 1H, J = 8.7, 2.3 Hz), 3.90 (s, 3H), 3.74 (s, 3H), 2.73 (d, 3H, J = 4.7 Hz), 1.36 (s, 9H), 0.87 (s, 9H), 0.02 (s, 3H), −0.11 (s, 3H); IR (neat) ν_{max} 3316, 2931, 2859, 1660, 1600, 1537, 1498, 1349, 1287, 1259, 1164 cm^{-1}; FABHRMS (NBA-NaI) *m*/*z* 930.2334 (M^+ + Na, $C_{39}H_{51}N_5O_{12}BrFSi$ requires 930.2369).

***N*-Methyl (2*S*,3*R*)-2-[*N*-[(*R*)-*N*-[(*R*)-*N*-[(*tert*-Butyloxy)-carbonyl]-(3,5-dihydroxy-4-methoxyphenyl)glycyl]-(3-bromo-4-methoxyphenyl)glycyl]amino]-3-hydroxy-3-(4-fluoro-3-nitrophenyl)propionamide (54).** A solution of **53** (16 mg, 0.017 mmol) in THF (5.3 mL) was treated with HOAc (1/100 v/v in THF, 5.5 μL, 0.096 mmol, 5.0 equiv) and Bu_4NF (1.0 M in THF, 116 μL, 0.116 mmol, 6.0 equiv) at 0 °C under Ar, warmed to 25 °C, and stirred for 1 h. The reaction mixture was quenched with the addition of saturated aqueous $NaHCO_3$ (5 mL) and extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with saturated aqueous NH_4Cl (2 × 5 mL), dried (Na_2SO_4), and concentrated in vacuo. PTLC (SiO_2, 10% $CH_3OH/CHCl_3$) afforded **54** (11.6 mg, 83%) as a pale yellow solid: $[\alpha]^{25}_D$ −10 (*c* 0.20, $CHCl_3$); ^{1}H NMR (acetone-d_6, 600 MHz) δ 8.01 (dd, 1H, J = 7.2, 2.0 Hz), 7.89 (d, 2H, J = 4.4 Hz), 7.79 (d, 2H, J = 8.9 Hz), 7.54 (m, 2H), 7.50 (d, 1H, J = 2.2 Hz), 7.47 (br s, 1H), 7.19 (dd, 1H, J = 8.5, 2.2 Hz), 7.12 (dd, 1H, J = 11.0, 8.7 Hz), 6.86 (d, 1H, J = 8.5 Hz), 6.49 (s, 2H), 6.17 (br s, 1H), 5.48 (d, 1H, J = 6.4 Hz), 5.41 (d, 1H, J = 2.2 Hz), 5.15 (d, 1H, J = 5.8 Hz), 4.66 (dd, 1H, J = 8.8, 2.4 Hz), 3.89 (s, 3H), 3.78 (s, 3H), 2.72 (d, 3H, J = 4.6 Hz), 1.37 (s, 9H); IR (neat) ν_{max} 3296, 2964, 1659, 1637, 1536, 1500, 1347, 1286, 1260, 1163, 1056 cm^{-1}; FABHRMS (NBA-NaI) *m*/*z* 816.1469 (M^+ + Na, $C_{33}H_{37}N_5O_{12}BrF$ requires 816.1504).

***N*-Methyl (*P*)- and (*M*)-(8*R*,11*R*,14*S*,15*R*)-11-(3-Bromo-4-methoxyphenyl)-8-[*N*-[(*tert*-butyloxy)carbonyl]amino]-5,15-dihydroxy-9,12-dioxo-4-methoxy-18-nitro-10,13-diaza-2-oxatricyclo[14.2.2.1]heneicosa-1(18),3(21),4,6,16,19-hexaene-14-carboxamide (55).** A solution of **54** (4.1 mg, 5.1 μmol) in anhydrous degassed DMSO (0.63 mL, 0.008 M) was treated with CsF (4.3 mg, 28 μmol, 5.6 equiv) at 25 °C under Ar and stirred for 11 h. The reaction mixture was filtered through Celite (EtOAc wash) and concentrated in vacuo. PTLC (SiO_2, 5% $CH_3OH/CHCl_3$) afforded (*P*)-**55** (1.6 mg, 41%) as a pale yellow solid, and (*M*)-**55** (2.1 mg, 54%) as a white solid.

For (*P*)-**55** (the less polar isomer): $[\alpha]^{25}_D$ −12 (*c* 0.065, $CHCl_3$); ^{1}H NMR (CD_3OD, 400 MHz) δ 8.05 (d, 1H, J = 2.1 Hz), 7.69 (d, 1H, J = 8.0 Hz), 7.48 (m, 1H), 7.37 (d, 1H, J = 8.6 Hz), 7.28 (d, 1H, J = 8.5 Hz), 7.10 (d, 1H, J = 8.6 Hz), 6.65 (m, 1H), 6.27 (m, 1H), 5.38 (s, 1H), 5.34 (d, 1H, J = 3.8 Hz), 5.15 (br s, 1H), 4.54 (d, 1H, J = 3.8 Hz), 3.96 (s, 3H), 3.91 (s, 3H), 2.77 (s, 3H), 1.43 (s, 9H); ^{1}H NMR (acetone-d_6, 600 MHz) δ 8.29 (s, 1H), 8.06 (d, 1H, J = 1.8 Hz), 7.79 (d, 1H, J = 9.0 Hz), 7.66−7.61 (m, 2H), 7.47 (m, 2H), 7.34 (d, 1H, J = 8.5 Hz), 7.11 (d, 1H, J = 8.5 Hz), 6.76 (s, 1H), 6.71 (m, 1H), 6.39 (s, 1H), 6.22 (m, 1H), 5.55 (dd, 1H, J = 5.6, 2.4 Hz), 5.47 (d, 1H, J = 8.0 Hz), 5.18 (d, 1H, J = 7.8 Hz), 5.12 (d, 1H, J = 4.9 Hz), 4.62 (d, 1H, J = 5.5 Hz), 3.94 (s, 3H), 3.91 (s, 3H), 2.73 (d, 3H, J = 4.6 Hz), 1.38 (s, 9H); IR (neat) ν_{max} 3304, 2978, 2926, 1652, 1575, 1532, 1505, 1456, 1436, 1349, 1258, 1164, 1086, 1022 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 906.0627 (M^+ + Cs, $C_{33}H_{36}N_5O_{12}Br$ requires 906.0598). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 25 °C) of (*P*)-**55** displayed the following diagnostic NOE cross-peaks: H-17/H-15 (s, δ 8.05/5.34), H-17/H-14 (s, δ 8.05/4.54), H-15/H-14 (s, δ 5.34/4.54), H-2′/H-11 (m, δ 7.48/5.38), H-6′/H-11 (s, δ 7.37/5.38), H-8/H-6 (s, δ 6.65/5.15).

For (*M*)-**55** (the more polar isomer): $[\alpha]^{25}_D$ −29 (*c* 0.12, $CHCl_3$); ^{1}H NMR (CD_3OD, 400 MHz) δ 8.21 (d, 1H, J = 1.6 Hz), 7.70 (d, 1H, J = 2.2 Hz), 7.68 (d, 1H, J = 2.1 Hz), 7.36 (d, 1H, J = 8.5 Hz), 7.31 (d, 1H, J = 8.4 Hz), 7.06 (d, 1H, J = 8.6 Hz), 6.60 (m, 1H), 5.99 (m, 1H), 5.37 (m, 2H), 5.17 (s, 1H), 4.64 (d, 1H, J = 3.0 Hz), 3.98 (s, 3H), 3.90 (s, 3H), 2.78 (s, 3H), 1.41 (s, 9H); ^{1}H NMR (acetone-d_6, 600 MHz) δ 8.33 (s, 2H), 7.68 (s, 3H), 7.43 (d, 1H, J = 8.6 Hz), 7.26 (d, 1H, J = 7.9 Hz), 7.05 (d, 1H, J = 8.2 Hz), 6.66 (s, 2H), 6.15 (m, 1H), 5.94 (m, 1H), 5.33 (s, 2H), 5.49 (d, 1H, J = 7.6 Hz), 5.31 (d, 1H, J = 8.8 Hz), 4.88 (m, 1H), 3.96 (s, 3H), 3.89 (s, 3H), 2.68 (s, 3H,) 1.39 (s, 9H); IR (neat) ν_{max} 3319, 2971, 2930, 1692, 1649, 1582, 1535, 1499, 1346, 1286, 1263, 1227, 1165, 1082, 1040 cm^{-1}; FABHRMS (NBA-CsI) *m*/*z* 906.0558 (M^+ + Cs, $C_{33}H_{36}N_5O_{12}Br$ requires 906.0598). The 2D ^{1}H−^{1}H ROESY NMR spectrum (CD_3OD, 600 MHz, 25 °C) of (*M*)-**55** displayed the following diagnostic NOE cross-peaks: H-20/H-15 (s, δ 7.67/5.38), H-20/H-14 (s, δ 7.67/4.65), H-14/H-15 (m, δ 4.65/5.38), H-6′/H-11 (w, δ 7.33/5.29), and H-8/H-6 (m, δ 5.15/6.62).

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Supporting Information Available: Experimental details and characterization of **10**, **11**, **13**, **14**, **16**, **18**−**21**, **23**−**24**, and **26**-**29**, the correlations of (*P*)-**30** with (*P*)-**28**, of **24** with **45**, of **30** with **37**, **33**-**36**, **44**-**45**, the conversion of (*P*)-**2** to (*P*)-**55**, **55**−**60**, and the preparation, cyclization, and correlation of the product atropisomers of the CO C14 hydroxymethyl derivative. Copies of the ^{1}H NMR of all compounds reported herein are also provided (77 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

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