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Synthesis of Iron Chelators. Enterobactin, Enantioenterobactin, and a Chiral Analogue

William H. Rastetter,*[1] Thomas J. Erickson, and Michael C. Venuti

Department of Chemistry, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139

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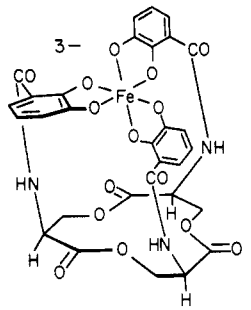
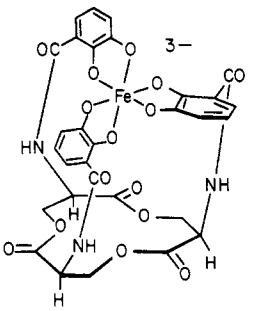
Enterobactin (3), the iron-binding ionophore of the enteric bacteria, has been synthesized from L-serine. The antipode of the natural product, enantioenterobactin (42), has been analogously assembled from D-serine, and the iron-binding and biological properties of the ligand have been investigated. Linear, chiral analogue 66 has been synthesized from L-asparagine, and the analogue Fe(III) complex 67 has been characterized. Attempts to assemble the trilactam analogue 4 have failed at the cyclization step.

To contend with the insolubility of ferric hydroxide in an aqueous milieu [$K_{sp}(Fe(OH)_3) = 10^{-38}$],[2] microbes have evolved specialized ligands or siderophores[3] to acquire and transport environmental iron into the cell. The siderophores bind Fe(III) by wrapping six ligating atoms around the metal center in either a right-handed (Δ) or left-handed (Λ) coordination propeller (see enantiomeric complexes 1 and 2). We have assumed that the chirality of an iron–

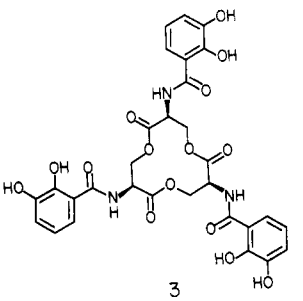


1, Δ-cis Fe(III)·enterobactin



2, Λ-cis Fe(III)·enantioenterobactin

siderophore complex may play a key role in the binding and transport of the complex into the microbial cell. To address the stereospecificity of the iron-gathering process in the enteric bacteria, we have synthesized the catechol-based siderophore enterobactin (3) and its antipode, enantioenterobactin (42, Scheme IV),[4] from L- and D-serine, respectively. Herein we report the details of these syntheses, including studies of racemization during the



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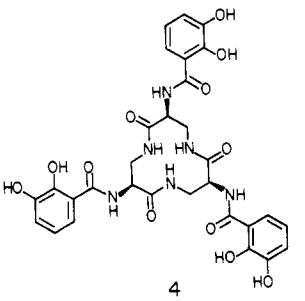

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coupling of serine derivatives. The biological activity of enantioenterobactin is discussed in light of the chirality of its iron complex. Also discussed are strategies and difficulties encountered in an attempted synthesis of the trilactam analogue of enterobactin, 4, which is of interest in the study of the intracellular release of iron from iron(III)–enterobactin. Intermediates from this synthesis have yielded the linear, chiral analogue 66 (Scheme VII); its synthesis and the characterization of the complex Fe(III)·66 (see 67) are reported.

Background. Enterobactin[5] is overproduced and excreted by *Escherichia coli* and related enteric bacteria under low iron stress. Solubilization of Fe(III) is achieved upon formation of the exceptionally stable octahedral complex 1 ($K_f = 10^{52}$).[6] An outer membrane receptor protein then functions in the binding and transport of the siderophore–iron complex into the bacterial cell.[7] In principle, two diastereomeric iron(III)–enterobactin complexes can exist, the right-handed or Δ-cis complex (1) and the left-handed or Λ-cis complex (not shown).[8] The in-

(1) Firmenich Assistant Professor of Natural Products Chemistry, Alfred P. Sloan Fellow, 1980–1982.
(2) Linke, W. F. "Solubilities. Inorganic and Metal-Organic Compounds", 4th ed.; Van Nostrand: Princeton, NJ, 1958; Vol. 1, p 1039.
(3) Raymond, K. N.; Carrano, C. J. *Acc. Chem. Res.* **1979**, *12*, 183 and references therein.
(4) Rastetter, W. H.; Erickson, T. J.; Venuti, M. C. *J. Org. Chem.* **1980**, *45*, 5011.
(5) (a) Pollack, J. R.; Neilands, J. B. *Biochem. Biophys. Res. Commun.* **1970**, *38*, 989. (b) O'Brien, I. G.; Gibson, F. *Biochim. Biophys. Acta* **1970**, *215*, 393.
(6) Harris, W. R.; Carrano, C. J.; Cooper, S. R.; Sofen, S. R.; Avdeef, A. E.; McArdle, J. V.; Raymond, K. N. *J. Am. Chem. Soc.* **1979**, *101*, 6097.
(7) Hollifield, W. C., Jr.; Neilands, J. B. *Biochemistry* **1978**, *17*, 1922.

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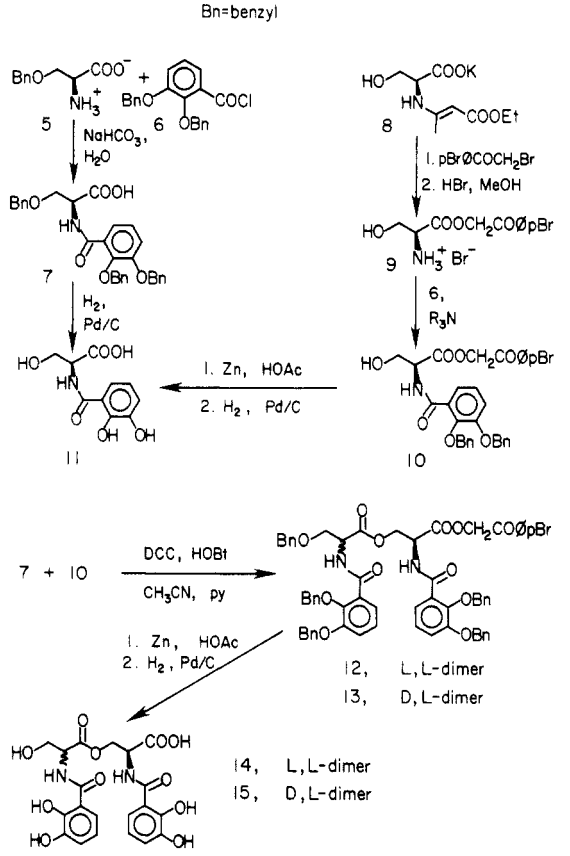
teraction of the outer membrane receptor protein with each diastereomer might be quite different. Were both Δ-*cis* and Λ-*cis* enterobactin complexes available and separable, the sensitivity of membrane binding and transport to changes in metal center configuration could be evaluated. However, on the basis of the chromatographic behavior of kinetically inert enterobactin metal complexes and from correlations of CD spectra, Raymond and co-workers[9] have concluded that ferric enterobactin exists in solution as the Δ-cis isomer to the exclusion of the diastereomeric Λ-cis complex. By analogy, ferric *enantio*enterobactin must exist as the Λ-cis isomer. Membrane receptor–siderophore complex interactions based on size, charge, and polarity must remain unchanged for iron(III)–enantioenterobactin. Bioassay of synthetic enantioenterobactin, thus, will reveal the dependence of binding and transport on solely the combination of platform and metal center chirality (natural, L-seryl and Δ-*cis*, **1**, vs. unnatural, D-seryl and Λ-*cis*, **2**).

Enterobactin is not reexcreted after the transport and iron-release processes;[3] i.e., a single ferric ion is transported by a molecule of enterobactin before the siderophore is hydrolyzed by the enzyme, enterobactin esterase (enterochelin esterase).[10] The hydrolysis step may be a prerequisite for iron release.[6,11] Alternatively, it has been suggested[12] that the hydrolysis of the ester bonds is only coincidently related to the iron-delivery step and that iron release from iron(III)–enterobactin is initiated by protonation and reduction of the complex to a neutral Fe(II) species.[13] Enterobactin analogue **4**, lacking the hydrolytically labile[14] seryl ester linkages of the natural product, might display some resistance to intracellular degradation by enterobactin esterase. Mutant *E. coli* cells deficient in the iron-release mechanism display a distinct pink coloration due to accumulated, highly colored iron(III)–enterobactin.[10b] Parent strain or mutant *E. coli* defective in the transport of iron(III)–enterobactin are white and do not accumulate the red complex. It is intriguing to wonder whether *E. coli* with an intact iron-release system, but unable to rapidly hydrolyze the amide bonds of Fe(III)·**4**, would also accumulate the complex and develop a pink, ferric catecholate coloration. Thus, bacterial studies with trilactam **4** might reveal information relating the enzymatic dismembering of the enterobactin backbone to the iron-delivery step.

Enantiomeric Enterobactins. Initial Approaches. The symmetry of enterobactin (**3**) dictates the synthetic approach to the siderophore; viz., an efficient synthesis will entail the coupling and subsequent cyclization of protected L-serine monomer units. Analogously, D-serine might serve as chiral precursor to enantioenterobactin (**42**). Within these broad outlines we have evaluated two fundamentally different approaches to enterobactin and its antipode. The approaches differ in the timing of the catechol ligand attachment to serine nitrogen. The first entails the coupling of differentially protected *N*-[2,3-bis(benzyloxy)benzoyl]serine monomers (for example **7** and **10**, Scheme I), an approach which obviates the need for multiple deprotections and acylations of serine nitrogen late in the synthetic sequence. The second utilizes urethane protection for the monomer amino groups (e.g., see Scheme IV) and introduces the *N*-benzoyl ligands only after cyclization of the seryl ester platform. The potential problems and the ultimate outcome of each approach are discussed below.

Scheme I



The risk inherent to any *N*-benzoyl monomer approach is the racemization of the activated ester required for monomer coupling. Nonetheless, the success achieved with dicyclohexylcarbodiimide/*N*-hydroxybenzotriazole (DCC/HOBt)[15] for the coupling of *N*-benzoyl amino acids[15e] suggested the use of these reagents for the coupling of protected *N*-(2,3-dihydroxybenzoyl)serine monomers. Scheme I shows the synthesis of the monomers and the results of the coupling experiments performed in the L-serine series.

Acylation of *O*-benzyl-L-serine (**5**) with 2,3-bis(benzyloxy)benzoyl chloride (**6**)[16] directly provides the alcohol-

(8) Were it possible to interchange the positions of the catecholate oxygens of one bidentate 2,3-dihydroxybenzoyl ligand in the Δ-cis complex **1**, the Δ-trans complex would be formed; the Λ-cis and Λ-trans complexes are related analogously. Dreiding molecular models clearly show, however, that the trans complexes are too strained to form.

(9) Raymond, K. N.; Kamal, A.-D.; Sofen, S. R. *ACS Symp. Ser.*, **1980**, *No. 119*, 133–167.

(10) (a) Greenwood, K. T.; Luke, R. K. J. *Biochim. Biophys. Acta* **1978**, *525*, 209. (b) Langman, L.; Young, I. G., Frost, G. E., Rosenberg, H.; Gibson, F. *J. Bacteriol.* **1972**, *112*, 1142.

(11) Cooper, S. R.; McArdle, J. V.; Raymond, K. N. *Proc. Natl. Acad. Sci. U.S.A.* **1978**, *75*, 3551.

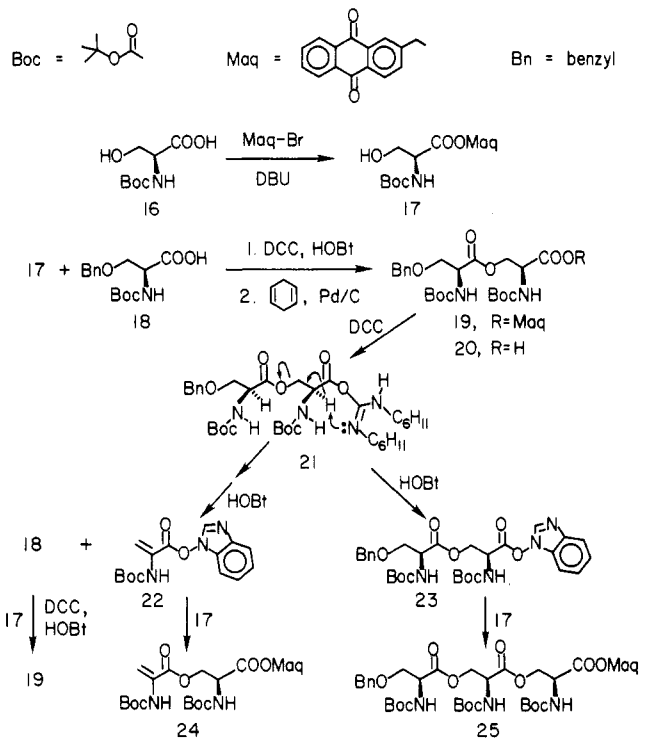
(12) The carbocyclic analogue of enterobactin, *cis*-1,5,8-tris(2,3-dihydroxybenzamido)cyclododecane (Corey, E. J.; Hurt, S. D. *Tetrahedron Lett.* **1977**, 3923), is remarkably effective in iron deliver delivery *E. coli*. On the basis of this observation, Hollifield and Neilands[7] have questioned the role of an esterase in the metabolic event(s) involved in the release of the coordinated ferric ion from iron(III)–enterobactin. The formation constant for the Fe(III) analogue complex, however, is not known, and, hence, the stability of the complex has not been directly compared to that of iron(III)–enterobactin ($K_f = 10^{52}$).[6]

(13) Hider, R. C.; Silver, J.; Neilands, J. B.; Morrison, I. E. G.; Rees, L. V. C. *FEBS Lett.* **1979**, *102*, 325.

(14) In aqueous buffer (Tris·HCl, 0.5 M, pH 8.0) enterobactin is reported to be hydrolyzed substantially after 3 h at 37 °C; after 20 h the predominant hydrolysis product is monomer **10** (Scheme I): O'Brien, I. G.; Cox, G. B.; Gibson, F. *Biochim. Biophys. Acta* **1970**, *201*, 453.

(15) (a) König, W.; Geiger, R. *Chem. Ber.* **1970**, *102*, 788. (b) Klausner, Y.; Chorev, M. *J. Chem. Soc., Chem. Commun.* **1975**, 973. (c) Chorev, M.; Knobler, Y.; Klausner, Y. *J. Chem. Res., Synop.* **1977**, 202. (d) Weber, U. *Z. Naturforsch., B: Anorg. Chem., Org. Chem.* **1976**, *31*, 1157. (e) Young, G. T. "Proceedings of the 12th European Peptide Symposium"; Hanson, H., Jakubke, H., Eds.; North-Holland Publishing Co.: Amsterdam, 1973; p 132.

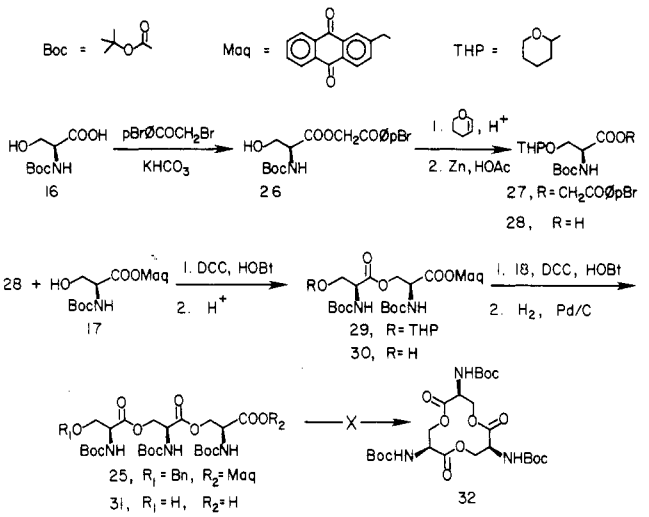
Scheme II



blocked monomer **7**. Acid-blocked monomer **10** is made from enamine **8**[17] by alkylation and deprotection (**8** → **9**) followed by acylation (**9** → **10**). Both monomers are readily deprotected to give the enterobactin monomer **11**.[14,18] Amino acid analysis[19] of serine derived from **11** by hydrolysis shows both protected monomers (**7** and **10**) to be of high chiral purity (>95% L). The coupling of monomers **7** and **10** with DCC/HOBt produces a 1:1 mixture of diastereomeric dimers **12** and **13**, separable by LC (silica gel). Fully deprotected dimers **14** and **15** show the chromatographic behavior (cellulose) of naturally derived dimer **14**.[14]

Racemization,[20] usually held in check by the DCC/HOBt procedure, becomes a rapid and undesired process with monomer acid **7**. Neither changes in solvent nor reaction conditions give any improvement in the 1:1 ratio of diastereomers formed in the coupling reaction. Apparently, the electron-releasing, *o*-benzyloxy group in monomer **7** promotes the nucleophilicity of the benzamide carbonyl oxygen and enhances the rate of azlactonization of the active ester formed from **7**. Other coupling methods lead either to azlactone formation from **7**[21a] or to dehydration of monomer alcohol **10**.[21b-d] In view of these early difficulties with the *N*-benzoyl monomer approach, it was abandoned in favor of a urethane-protection route.

Scheme III



Urethane protection for serine nitrogen avoids the problem of racemization[20] during seryl ester formation.[22] Inherent to this approach is urethane deprotection and introduction of *N*-benzoyl ligands after the complete assembly of the seryl ester platform. The required N-acylation of the three amino groups in the cyclic triester of L- or D-serine (see **40**, Scheme IV), however, poses a potential risk late in the synthetic sequence owing to the propensity for *O*- to *N*-acyl shifts in *O*-acylserine derivatives.[23] This risk could be evaluated only through synthesis of the planned intermediate, cyclic *O*-acylserine derivative **40**.

Schemes II and III show initial, yet ultimately unsuccessful, urethane approaches to enterobactin. As shown in Scheme II, monomer alcohol **17** is formed (94%) upon alkylation of *N*-(*tert*-butyloxy)carbonyl-L-serine[25] with 2-(bromomethyl)anthraquinone[26] (MaqBr). Coupling of **17** with *O*-benzyl-*N*-[(*tert*-butyloxy)carbonyl]-L-serine (**18**)[27] by the DCC/HOBt procedure produces crystalline dimer **19** in 80% yield.[22] Catalytic transfer hydrogenolysis[28] selectively and quantitatively removes the Maq ester, producing acid dimer **20**. Despite the successful coupling of monomers, the coupling of acid dimer **20** with monomer alcohol **17** is not straightforward. The coupling produces a low yield of crystalline trimer **25** (19%), as well as crystalline dehydro dimer **24** (7%) and fully protected crystalline dimer **19** (35%). As shown in Scheme II, these three products must arise via active ester **21** which can either couple (**21** → **23** → **25**) or fragment (**21** → **18** + **22**). The unexpected dimeric products **19** and **24** arise by coupling of the fragments (**18** and **22**) with monomer alcohol **17**.

For avoidance of dimer fragmentation, serine oligomerization must be achieved by deprotection and chain extension at the seryl hydroxyl group (cf. carboxyl de-

(16) Venuti, M. C.; Rastetter, W. H.; Neilands, J. B. *J. Med. Chem.* **1979**, *22*, 123.

(17) (a) Balog, A.; Breazu, D.; Daicoviciu, C.; Varga, E.; Beu, L.; Gonczy, F. *Rev. Roum. Chim.* **1971**, *16*, 1601. (b) Maclaren, J. A. *Aust. J. Chem.* **1972**, *25*, 1293.

(18) O'Brien, I. G.; Cox, G. B.; Gibson, F. *Biochim. Biophys. Acta* **1969**, *177*, 321.

(19) Diastereomeric dipeptide analysis performed according to: Manning, J. M.; Moore, S. *J. Biol. Chem.* **1968**, *243*, 5591. We thank Drs. Ralph Hirschmann and Carl Bennett of Merck Sharp and Dohme Research Laboratories, West Point, PA, for carrying out these analyses.

(20) Review: Kemp, D. S. In "The Peptides"; Gross, E., Meienhofer, J., Eds.; Academic Press: New York, 1979; Chapter 7.

(21) (a) DCC, pyridine. (b) *N*-Methyl-2-chloropyridinium iodide, triethylamine: Mukaiyama, T.; Usui, M.; Shimada, E.; Saigo, K. *Chem. Lett.* **1975**, 1045. (c) Ethyl diazodicarboxylate, triphenylphosphine: Mitsunobu, O.; Eguchi, M. *Bull. Chem. Soc. Jpn.* **1971**, *44*, 3427. (d) *N,N'*-Carbonyldiimidazole: Staab, H. A.; Mannschreck, A. *Chem. Ber.* **1962**, *95*, 1284.

(22) Dimer **19** (Scheme II) was compared (TLC, ^{1}H NMR) with a diastereomeric dimer produced from *racemic* monomer acid **16** and optically pure L monomer **17**. TLC and ^{1}H NMR showed no detectable diastereomeric dimer in the product from coupling of L-serine derived **16** and **17**.

(23) Schröder, E.; Lübke, K. "The Peptides"; Academic Press: New York, 1965; Vol. 1, pp 213–214.

(24) The viability of the urethane approach was first shown by Corey in his enterobactin synthesis: Corey, E. J.; Bhattacharyya, S. *Tetrahedron Lett.* **1977**, 3919.

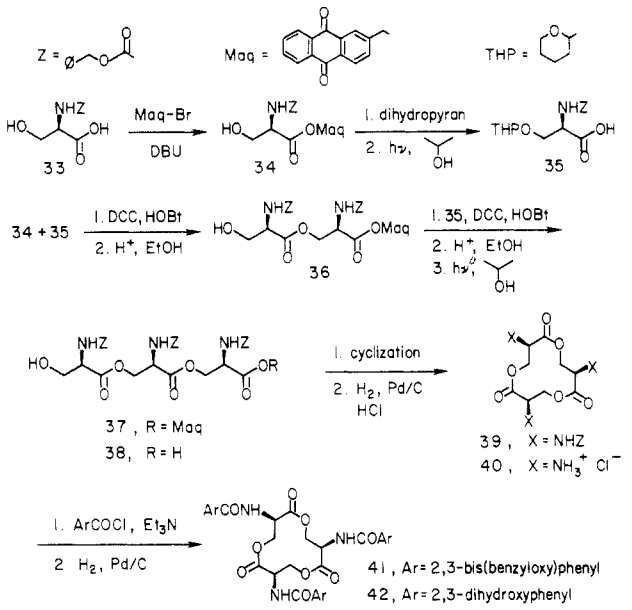
(25) "Methoden der Organischen Chemie (Houben-Weyl)"; Müller, E., Ed.; Georg Thieme Verlag: Stuttgart, 1974; Band Teil I, p 132.

(26) Kemp, D. S.; Reczek, J. *Tetrahedron Lett.* **1977**, 1031.

(27) Wuench, E.; Zwick, A. *Chem. Ber.* **1964**, *97*, 2497.

(28) Baer, E.; Maurukas, J. *J. Biol. Chem.* **1955**, *212*, 25.

Scheme IV



protection and extension, Scheme II). Thus, monomer **28**, made as shown in Scheme III (overall 76%), can be coupled with alcohol monomer **17**, cleanly producing dimer **29**. Deprotection yields crystalline alcohol dimer **30** (overall 80% from **17**). Coupling of **30** with *O*-benzyl-blocked monomer **18** (from Scheme II) gives the previously obtained (Scheme II) protected trimer **25** as a crystalline solid (66%). Hydrogenolysis achieves deprotection of both ends of trimer **25**, yielding hydroxy acid **31** (100%). Ultimately, (*tert*-butyloxy)carbonyl-protected trimer hydroxy acid **31** failed to cyclize cleanly under a variety of conditions, including those used by Corey et al.[24] for cyclization of the corresponding *N*-[(benzyloxy)carbonyl]-protected trimer hydroxy acid (see enantiomer **38**, Scheme IV). Crude cyclization mixtures derived from trimer **31** could not be purified nor deprotected to give a cyclic triammonium salt (see enantiomer **40**, Scheme IV).

Syntheses of the Enterobactins. Our successful approach to the enterobactins is outlined in Scheme IV for the D-seryl series. *N*-[(Benzyloxy)carbonyl]-D-serine[28] (**33**) is alkylated with 2-(bromomethyl)anthraquinone[26] (MaqBr), giving acid-protected monomer **34** (92%). Hydroxyl-protected monomer **35** is derived from **34** by reaction with dihydropyran followed by photoreductive deprotection[26] of the Maq ester (82% overall). Coupling of **34** and **35** and removal of the THP protective group yields crystalline dimer alcohol **36** (88% overall). Further coupling of **36** with **35** and THP removal yields crystalline trimer alcohol **37** (95% overall). Removal of the Maq ester generates the enantiomer (**38**) of Corey's linear trimer enterobactin precursor[24] (67–82%).

Trimer **38** can be cyclized in low yield by DCC/HOBt coupling, by Masamune's *tert*-butyl thioester/cuprous triflate method[29] or, preferably, by Corey's imidazolyl disulfide procedure.[24] Hydrogenolysis of crystalline **39** in the presence of HCl proceeds smoothly to trihydrochloride salt **40**[24] which is acylated without isolation to give hexabenzylenantioenterobactin (**41**, 61–69% overall). The successful triacylation **40** → **41** and the similar conversion previously reported by Corey et al.[24] dispelled our fears concerning *O*- to *N*-acyl shifts in the triamine liberated from salt **40**. Hydrogenolysis of **41** yields enantioenterobactin (**41**, 70–90%) which is indistinguishable from natural enterobactin by ^{1}H NMR, IR, TLC, UV, field-desorption mass spectrometry (M^+, m/e 669), and melting point. Ferric enantioenterobactin (**2**) displays a CD spectrum which is the mirror image of that measured[30] for the natural ferric complex (**1**, Figure 1).

(29) Masamune, S.; Hayase, Y.; Schilling, W.; Chan, W. K.; Bates, G. S. *J. Am. Chem. Soc.* **1977**, *99*, 6756.

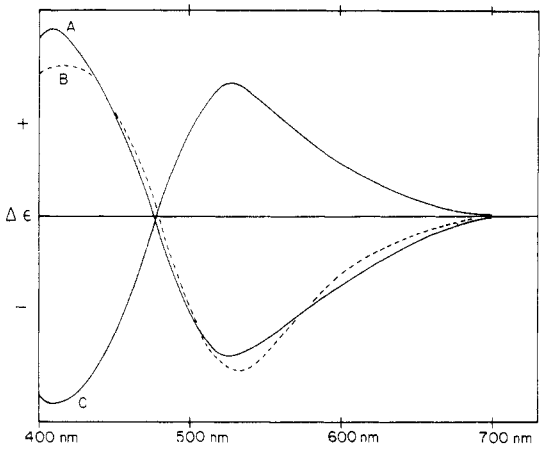
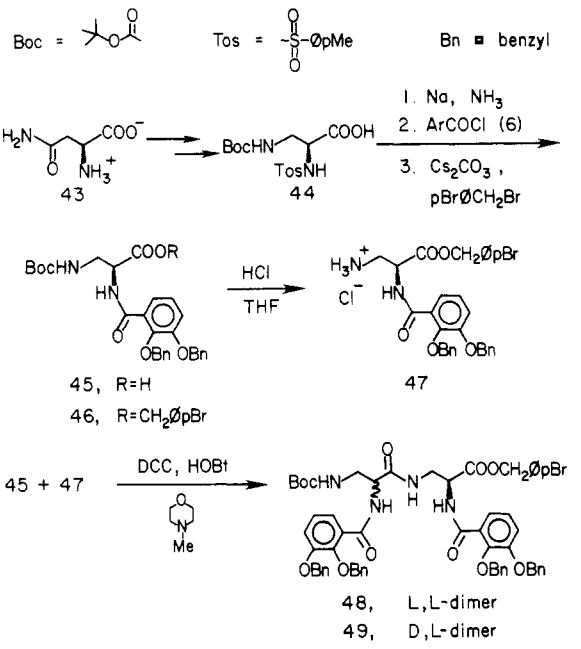


Figure 1. Circular dichroism spectra: A, iron(III)–enterobactin; B, ferric complex **67**; C, iron(III)–enantioenterobactin.

Scheme V



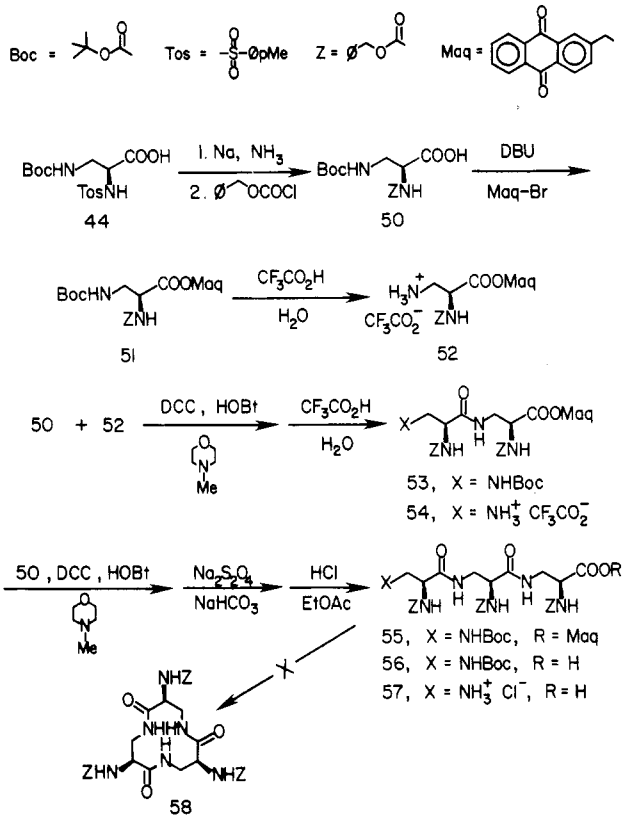
Scheme IV applied to *N*-[(benzyloxy)carbonyl]-L-serine[28] yields enterobactin indistinguishable in all respects (vide supra) from natural material.

Amide Analogues. Approaches to Trilactam 4. In place of serine a chiral source of α,β-diaminopropionic acid is required to adapt our enterobactin approaches to the assembly of analogue **4**. L-Asparagine (**43**, Scheme V) fills this role and provides a convenient way of differentiating the α- and β-amino groups of monomeric intermediates. Thus, literature procedures[31] convert asparagine (**43**) into

(30) (a) Salama, S.; Strong, J. D.; Neilands, J. B.; Spiro, T. G. *Biochemistry* 1978, *17*, 3781. (b) Rogers, H. J.; Synge, C.; Kimbar, B.; Bayley, P. M. *Biochim. Biophys. Acta* **1977**, *497*, 548.

(31) (a) Ressler, C. *J. Am. Chem. Soc.* **1960**, *82*, 1641. (b) Yoneda, N.; Fujii, T.; Uneda, M.; Yasuo, H.; Taguchi, Y.; Okurnura, K. *Yakugaku Zasshi* **1969**, *89*, 98; *Chem. Abstr.* **1969**, *70*, 88228. (c) Pozdnev, V. F. *Chem. Natl. Compd. (Engl. Transl.)* **1974**, *10*, 782. (d) Hallett, A.; Wuensch, E.; Keller, O.; Wersin, G. *Hoppe-Seyler's Z. Physiol. Chem.* **1976**, *357*, 1651.

Scheme VI



monomer 44[32] via tosylation, Hofmann rearrangement, and (*tert*-butyloxy)carbonyl protection of the β-nitrogen. α deprotection of 44 by reductive cleavage of the tosyl sulfonamide allows the introduction of the *N*-[2,3-bis(benzyloxy)benzoyl] group or of urethane protective groups at the α-nitrogen. The former option allows a further test of racemization during the coupling of monomers bearing the protected 2,3-dihydroxybenzoyl ligand (cf. Scheme I). The syntheses and coupling of the *N*-benzoyl monomers is shown in Scheme V.

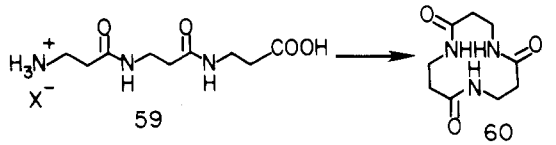
Acid monomer **45** is produced (84%) upon α deprotection of **44** and acylation of the α-amine with acid chloride **6**. From **45**, monomer amine salt **47** is formed upon carboxylate alkylation (**45** → **46**, 98%) and β deprotection (**46** → **47**, 84%). The DCC/HOBt-mediated coupling of **45** and **47** affords a 1:1 mixture of diastereomeric dimers **48** and **49** in quantitative yield. The amine-active ester coupling is slow relative to active ester racemization. Here, as in the serine acid monomer (**7**, Scheme I), the *N*-[bis(benzyloxy)benzoyl] group apparently promotes azlactonization and the loss of monomer chiral integrity. The composition of the mixture **48**/**49** is verified upon comparison with authentic diastereomeric dimer made from *racemic* acid monomer (cf. **45**) and monomer amine salt **47**.

The success of the (benzyloxy)carbonyl protective group for serine nitrogen in the enterobactin synthesis (Scheme IV) prompted the synthesis of linear trimer amino acid **57** (Scheme VI). (Benzyloxy)carbonyl-protected trimer **57**, thus, is analogous to the enantioenterobactin linear precursor **38**. Cyclization of **57** would afford the trilactam platform (see **58**) required for analogue **4**. The monomer syntheses and coupling steps for the urethane route are shown in Scheme VI.

(32) Moore, S.; Patel, R. P.; Atherton, E.; Kondo, M.; Meienhofer, J. *J. Med. Chem.* **1976**, *19*, 766.

Known acid monomer **50**[32] is made from the L-asparagine-derived, chiral precursor **44**. The amine monomer **52** is produced from **50** by alkylation (**50** → **51**, 87%) and β deprotection (**51** → **52**, 87%). Coupling of monomers and amine deprotection affords crystalline dimer amine salt **54** (87% overall). Further coupling yields trimer **55**; reductive cleavage of the Maq ester leads to crystalline trimer acid **56** (85% overall). Finally, cleavage of the terminal (*tert*-butyloxy)carbonyl protective group provides the desired trimer amino acid **57** as a white, crystalline solid.

To examine procedures which might be applied to the cyclization of trimer **57**, we performed closure reactions with model trimer **59**. Tri-β-alanine hydrochloride (**59**)[33]



cyclizes in low yield to cyclo(tri-β-alanine) (**60**)[34] with DCC/HOBt (8%) or diphenylphosphoryl azide/triethylamine[35] (14%). Application of these procedures to trimer **57** produces only polymeric material, even at high dilution. Cyclizations with *o*-phenylene phosphorochloridite[36] or via the *p*-nitrophenyl ester[37] also fail to produce the desired cyclic material (**58**). Sterically, active esters from linear trimer **57** resemble active esters from enterobactin precursor **38** (Scheme IV). The failure to achieve the trilactam cyclization may reflect conformational rather than steric differences between the ester and amide precursors. It is interesting to note that a Dreiding or Corey–Pauling–Koltun (CPK) model of trilactam **58** is easily constructed with all three amide linkages in the trans conformation. Thus, even the trans,trans conformation of linear diamide **57** should be able to cyclize. Nonetheless, intermolecular condensation (polymerization) proceeds faster than cyclization of trimer **57**.

Despite the inability to cyclize trimer **57**, an analogue of enterobactin might be built upon the *linear* backbone of the trimer. Introduction of 2,3-dihydroxybenzoyl ligands at the α-nitrogens (see **66**, Scheme VII) and chelation of Fe(III) might yield a complex geometrically very similar to iron(III)–enterobactin (**1**). The iron center of the complex presumably would hold the ends of the linear backbone in close proximity, forming a nearly circular array (see **67**). Further, the chirality of the backbone, as in enterobactin, might influence the chirality of the iron center. The transmission of chiral information from backbone to metal center, in principle, might be very different for the linear analogue with its different flexibility and greater degrees of freedom. The synthesis of the linear analogue (**66**) is shown in Scheme VII.

Trimer acid **56** (from Scheme VI) is converted into trimer ester **61** by the action of diazomethane; hydrogenolysis and acylation with 2,3-bis(benzyloxy)benzoyl chloride (**6**) affords fully protected trimer **63** (54% overall). Terminal amine deprotection (91%) and acetylation (72%) provides hexabenzyl-protected trimer **65**. Hydrogenolysis of **65** yields the desired analogue **66** (75%).

(33) Adams, E.; Davis, N. C.; Smith, E. L. *J. Biol. Chem.* **1952**, *199*, 845.

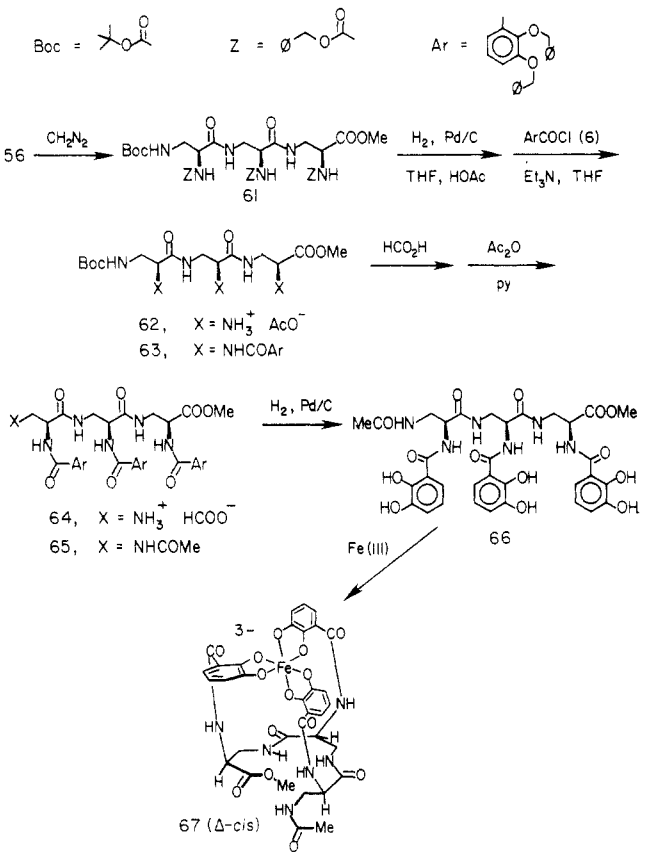
(34) (a) Rothe, M. *Acta Chim. Acad. Sci. Hung.* **1959**, *18*, 449. (b) Rothe, M.; Rothe, I.; Brung, H.; Schwenke, K. D. *Angew. Chem.* **1959**, *71*, 700.

(35) Brady, S. F.; Varga, S. L.; Freidinger, R. M.; Schwenk, D. A.; Mendlowski, M.; Holly, F. W.; Veber, D. R. *J. Org. Chem.* **1979**, *44*, 3101.

(36) Rothe, M.; Kreiss, W. *Angew. Chem., Int. Ed.* **1973**, *12*, 1012.

(37) Bohman-Lindren, G.; Ragnarsson, U. *Tetrahedron* **1972**, *28*, 4631.

Scheme VII



Fe(III) complex **67** forms upon mixing **66** and $FeCl_3$ in aqueous methanolic buffer at pH 7.2. The shape, sign, and intensity of the CD curve for **67** are nearly identical with that observed for iron(III)–enterobactin (Figure 1), indicating the Δ-cis geometry for the linear analogue complex (**67**). The formation constant for complex **67** has been measured at pH 7.2 by a competition experiment using ethylenediaminetetraacetic acid (EDTA) as an Fe(III) scavenger. The absorbance of red complex **67** (λ_{max} 495 nm) is 4%, 9%, and 86%, diminished at **67**/EDTA ratios of 1:10, 1:100, and 1:1000, respectively. From these data is calculated a formation constant for **67**, $K_f = 10^{46.5}$. Analogue complex **67** is, thus, some 5–6 orders of magnitude less stable than the natural complex **1** ($K_f = 10^{52}$).[6]

Iron-Transport Studies. Synthetic enterobactin (**3**), derived from L-serine, shows the full biological activity of naturally produced material.[38,42] Thus, synthetic **3** supports growth of mutant RW193 of *E. coli* K-12[39] which is deficient in the biosynthesis of the natural siderophore. Enantioenterobactin (**42**), in contrast, does not support bacterial growth; rather, **42** shows inhibitory activity toward strain RW193.[42] Fe(III), tightly sequestered in the form of D-seryl Λ-cis complex **2**, is unavailable to the bacterial transport system. The presence of siderophore antipode **42** in the growth medium thereby effectively prevents bacterial growth through iron starvation. This result is more dramatic than that observed recently in transport studies with enantioferrichrome,[40] the antipode of a hydroxamate siderophore of fungal origin. The uptake of ^{55}Fe(III)·enantioferrichrome is roughly 25–50% as efficient as the uptake of the natural siderophore complex.

The transport study with enantioenterobactin (**42**) shows high chiral specificity of the outer membrane receptor protein for the natural siderophore complex (**1**) having an L-seryl platform and a Δ-cis metal center. The membrane chiral specificity for the natural complex may be determined by the chirality of the seryl triester backbone, by the chirality of the metal center, or by both of these factors. The large diameter of the chiral iron(III)–catecholate center (~14 Å) and the insensitivity of the membrane transport process to changes in the platform[16] suggest that the configuration of the metal center is the more important factor in determining membrane recognition. In further support of this, a synthetic, achiral, carbocyclic analogue of enterobactin[12] forms a racemic Fe(III) complex (50% Δ-cis and 50% Λ-cis) which is almost exactly *half* as effective as ferric enterobactin in competition for the solubilized outer membrane receptor of *E. coli*.[7] This observation is consistent with membrane receptor binding of only the Δ-cis complex, assuming slow equilibration of the two antipodes under conditions of the experiment.[41]

Experimental Section

General Methods. All reactions were performed under a nitrogen atmosphere unless otherwise noted. Photolyses were carried out in Pyrex glassware by using a Rayonet-Srinivasan Griffin photochemical reactor (Southern New England Ultraviolet Co.) using RPR 3500-Å lamps. Hydrogenations at 40–50 psi of H_2 were performed in a Parr apparatus.

Melting points are uncorrected and were determined in open-ended capillary tubes on a Mel-Temp or Thomas-Hoover melting point apparatus. ^{1}H NMR spectra at 60 MHz were recorded on either a Varian T-60, Perkin-Elmer R-24B, or JEOL FX60 Q instrument; 90-MHz ^{1}H NMR spectra were obtained on a JEOL FX90 Q and 250-MHz ^{1}H NMR spectra on a Bruker WM-250 spectrometer. Chemical shifts are reported on the δ scale relative to tetramethylsilane. Infrared spectra were recorded on either a Perkin-Elmer 567 or Perkin-Elmer 283B spectrometer. Circular dichroism (CD) spectra were recorded on a Cary 60 spectrometer, and optical rotations measured with a Perkin-Elmer 141 polarimeter. Ultraviolet and visible spectra were recorded on either a Perkin-Elmer 554 or Perkin-Elmer 330 spectrophotometer. Low-resolution mass spectra were determined on a Varian MAT 44 and high-resolution mass spectra on a CEC 110B Mattauch-Herzog (Du Pont Instruments) mass spectrometer. Field-desorption mass spectra were determined on a Varian MAT 731 by using carbon emitters.

Analytical TLC was carried out on E. Merck 0.2-mm F-254 silica gel on plastic backing, E. Merck 0.25-mm F-254 silica gel on glass plates, or Analtech 0.25-mm GF silica gel on glass plates. Compounds were detected by using UV, iodine vapor, or 7% ethanolic phosphomolybdic acid char. Analytical chromatograms of enterobactin and other catechol-containing analogues were developed on Eastman cellulose on aluminum plates and visualized with 3% ferric chloride in 1 N HCl(aq). Preparative thin-layer chromatography was performed on E. Merck silica gel F-254 plates (20 cm × 20 cm × 2.5 mm). Gravity column chromatography was carried out on E. Merck silica gel 60 (70–230 mesh), and flash chromatography was performed according to Still[43] on E. Merck silica gel 60 (230–400 mesh). Liquid chromatography (LC) was carried out on a Waters ALC/GPC 204 instrument using a μ-Porasil analytical column and UV detection (254 nm) or on a Waters Prep-500 instrument using dual silica gel cartridges.

Benzene, toluene, *N*,*N*-dimethylformamide, pyridine, acetonitrile, and CH_2Cl_2 were statically dried over 4-Å molecular sieves. *N*-Methylmorpholine was distilled from BaO or dried over 4-Å molecular sieves; Et_3N was distilled from CaH_2. THF was distilled from Na/PhCOPh ketyl; DCC was distilled in vacuo. Anhydrous Et_2O (Mallinckrodt) was used from sealed cans. 2,3-Di-

(38) Details of the biological studies are discussed by: Neilands, J. B.; Erickson, T. J.; Rastetter, W. H. *J. Biol. Chem.* **1981**, *256*, 3831.
(39) American Type Culture Collection, No. 33475.
(40) Winkelman, G. *FEBS Lett.* **1979**, *97*, 43.
(41) The rate of racemization of the kinetically inert Fe(III) complex (see ref 9) is not known.
(42) We thank Professor J. B. Neilands (Berkeley) for testing of the enantiomeric enterobactins.
(43) Still, W. C.; Kohn, M.; Mitra, A. *J. Org. Chem.* **1978**, *43*, 2923.

hydroxybenzaldehyde and *O*-benzyl-L-serine were obtained from Sigma Chemical Co.

The following abbreviations are used: DMF = *N,N*-dimethylformamide, DCC = *N,N'*-dicyclohexylcarbodiimide, DCU = *N,N'*-dicyclohexylurea, HOBt = *N*-hydroxybenzotriazole, 18-crown-6 = 1,4,7,10,13,16-hexaoxacyclooctadecane, DBU = 1,8-diazabicyclo[5.4.0]undec-7-ene, THF = tetrahydrofuran, MaqBr = 2-(bromomethyl)anthraquinone, Boc = (*tert*-butyloxy)carbonyl, Me_2SO = dimethyl sulfoxide.

2,3-Bis(benzyloxy)benzoyl Chloride (6). To a solution of 2,3-dihydroxybenzaldehyde (50.0 g, 362 mmol) and benzyl chloride (100 mL, 868 mmol) in absolute EtOH (450 mL) was added K_2CO_3 (62.5 g, 452 mmol). The mixture was refluxed 18 h; removal of solvent in vacuo and extractive workup (EtOAc/H_2O) provided crude 2,3-bis(benzyloxy)benzaldehyde as a moist solid: TLC (silica gel, 3:1 CH_2Cl_2–EtOAc) R_f 0.72.

The crude aldehyde (ca. 362 mmol) was dissolved in acetone (350 mL) and the solution diluted with H_2O (350 mL). To the cloudy mixture were added sulfamic acid (H_2NSO_3H, 49.2 g, 507 mmol) and sodium chlorite (80% $NaClO_2$, 43.0 g, 380 mmol) in portions over 30 min.[44] The resulting solution was stirred 1 h at ambient temperature in an open vessel. Removal of acetone in vacuo yielded crude 2,3-bis(benzyloxy)benzoic acid as a precipitate in the remaining aqueous mixture. The solid was collected, washed with absolute EtOH, dried, and recrystallized from aqueous EtOH to yield pure 2,3-bis(benzyloxy)benzoic acid: 113.4 g (94%); mp 124–124.5 °C (lit.[45] mp 124 °C); ^{1}H NMR (60 MHz, acetone-d_6) 10.5 (s, 1 H), 7.05–7.75 (m, 13 H), 5.28 (s, 2 H), 5.25 (s, 2 H); IR (KBr) 3030 (br), 1688, 1572, 1470, 1450, 1410, 1375, 1300, 1255, 1215, 1030 cm^{-1}; mass spectrum, *m/e* 344 (M^+), 243 ($M^+ - C_7H_7$), 225 (243 – H_2O), 91 ($C_7H_7^+$).

To a suspension of the acid (3.34 g, 10.0 mmol) in dry benzene (20 mL) at 5 °C was added DMF (3 drops) followed by oxalyl chloride (3.5 mL, 40 mmol) dropwise. The resulting solution was stirred 30 min and then evaporated in vacuo to an oil. Generally the oil was used directly for acylations, but the acid chloride could be crystallized by trituration with 20:1 petroleum ether–Et_2O, yielding 3.33 g (94%) of **6**: mp 55–56 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 7.1–7.5 (m, 13 H), 5.1 (s, 4 H); IR (KBr) 1775, 1685, 1570, 1450, 1375, 1310, 1265, 1225 cm^{-1}; mass spectrum, *m/e* 352 (M^+), 317 ($M^+ - {}^{35}Cl$), 289 ($M^+ - CO^{35}Cl$), 261 ($M^+ - C_7H_7$), 91 ($C_7H_7^+$).

***N*-[2,3-Bis(benzyloxy)benzoyl] Monomer Acid 7.** To a chilled solution of *O*-benzyl-L-serine (**5**; 10.0 g, 51.3 mmol) and NaOH (4.10 g, 102.5 mmol) in H_2O (100 mL) was added dropwise a solution of acid chloride **6** (17.6 g, 50.0 mmol) in THF (30 mL). The mixture was stirred 1 h at 0 °C and 2 h at ambient temperature and then acidified to pH 2 with concentrated aqueous HCl. Extractive workup (EtOAc/H_2O) and recrystallization from EtOAc yielded 20.0 g (78%) of monomer acid **7**: mp 121 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 9.25 (br s, 1 H), 9.05 (d, 1 H), 6.90–7.85 (m, 18 H), 5.18 (s, 4 H), 5.00 (m, 1 H), 4.42 (s, 2 H), 3.85 (m, 2 H); IR (KBr) 3360, 3050 (br), 1755, 1635, 1575, 1538, 1455, 1265, 1195, 1140, 1105, 1052, 1040, 960 cm^{-1}; mass spectrum, *m/e* 511 (M^+), 420 ($M^+ - C_7H_7$), 402 (420 – H_2O), 358 (402 – CO_2), 91 ($C_7H_7^+$).

Monomer acid **7** (1.10 g, 2.15 mmol) was fully deprotected by hydrogenolysis over 10% Pd on carbon (110 mg) in EtOH–HOAc (20:1, 10 mL). Filtration of catalyst, evaporation of solvent, and extractive workup [EtOAc/HCl(5% aq), NaCl(aq) wash] yielded monomer **11** (0.480 g, 93%) as an off-white powder displaying $[\alpha]_D$ +31.2° (*c* 2, MeOH). Hydrolysis of the monomer to give serine was achieved over 20 h at 110 °C in 6 N HCl(aq). Coupling of the serine so produced with L-leucine-*N*-carboxyanhydride and amino acid analysis of the resulting diastereomeric dipeptides[19] showed the serine to be 4.1% D and 95.9% L. L-Serine subjected to the same hydrolysis conditions analyzed for 1.2% D-serine. On adjustment for serine racemization during hydrolysis, the chiral purity of monomer **11** (monomer **7**) thus was ≥97.1% L [100% – (4.1% – 1.2%)].

***N*-[2,3-Bis(benzyloxy)benzoyl] Monomer Alcohol 10.** A modification of the literature procedure[17] was used. L-Serine ethyl acetoacetate enamine potassium salt **8** (mp 175–177 °C; 20.3 g, 79.5 mmol) was suspended in CH_3CN (750 mL), and to the mixture were added *p*-bromophenacyl bromide (22.1 g, 79.5 mmol) and 18-crown-6 (1.0 g, 3.8 mmol). The suspension was refluxed 30 min, cooled to ambient temperature and filtered to remove precipitated salts. The filtrate was evaporated, and the solid residue was dissolved in freshly prepared HBr–MeOH (100 mL, saturated) and stirred 10 min. The dark mixture was quickly evaporated to a solid residue and recrystallized from 2-propyl alcohol to yield hydrobromide salt **9** (29.0 g, 95%) as a tan solid: mp 162–163 °C; ^{1}H NMR (60 MHz, D_2O) 7.43 (AB q, 4 H), 5.50 (s, 2 H), 4.72 (br s, HOD), 4.50 (t, 1 H), 4.12 (d, 2 H); IR (KBr) 3385, 3220, 2905, 1740, 1690, 1580, 1510, 1430, 1400, 1365, 1272, 1215, 1105, 1065, 1045, 1005, 962 cm^{-1}.

To a stirred solution of hydrobromide salt **9** (20.0 g, 52 mmol) and diisopropylethylamine (27 mL, 155 mmol) in dry THF (200 mL) was added dropwise a solution of acid chloride **6** (17.9 g, 50.9 mmol) in THF (ca. 30 mL). The mixture was stirred at ambient temperature for 2 h and then concentrated in vacuo to one-quarter volume. Extractive workup [EtOAc/5% HCl(aq), 5% $NaHCO_3$(aq) and NaCl(sat,aq) washes] and recrystallization from EtOAc yielded monomer alcohol **10**: 21.20 g (60%); mp 117–118 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 9.00 (d, 1 H), 7.0–7.9 (m, 17 H), 6.20 (br s, 1 H), 5.30 (AB q, 2 H), 5.13 (2 s, 4 H), 5.0 (m, 1 H), 4.05 (m, 2 H); IR (KBr) 3470 (br), 3360, 2915, 1752, 1700, 1655, 1585, 1572, 1520, 1450, 1372, 1260, 1190, 1070, 965 cm^{-1}; mass spectrum, *m/e* 617 and 619 (M^+, ^{79}Br and ^{81}Br), 526 (M^+ – Br), 420 ($M^+ - C_8H_6BrO$).

Monomer alcohol **10** was fully deprotected by Zn/HOAc reduction followed by hydrogenolysis. Thus, alcohol **10** (1.30 g, 2.10 mmol) in glacial acetic acid was stirred with zinc powder (5.0 g, 77 mmol) for 2 h. The zinc was removed by filtration and the filtrate evaporated in vacuo. Acid–base extractive purification of the crude product [Et_2O/5% $NaHCO_3$(aq); aqueous layer washed with Et_2O, acidified to pH 2 with concentrated HCl, and extracted with EtOAc; organic layer washed with 5% HCl(aq) and NaCl(sat, aq)] and hydrogenolysis (conditions as for **7**, vide supra) gave monomer **11** (0.350 g, 70%) as an off-white powder displaying $[\alpha]_D$ +30.5° (*c* 2, MeOH). Serine derived from **11** (**10**) was 5.6% D and 94.4% L. On adjustment for serine racemization during hydrolysis (vide supra), the chiral purity of monomer **11** (monomer **10**) thus was ≥95.6% L [100% – (5.6% – 1.2%)].

Diastereomeric *N*-[2,3-Bis(benzyloxy)benzoyl] Dimers 12 and 13. To a solution of acid monomer **7** (5.11 g, 10.0 mmol), alcohol monomer **10** (5.56 g, 9.0 mmol), and HOBt (2.70 g, 17.6 mmol) in dry CH_3CN at 0 °C was added DCC (2.60 g, 12.6 mmol). The mixture was stirred 1 h at 0 °C and 1 h at ambient temperature; pyridine (1.7 mL, 21 mmol) was added and the mixture stirred 48 h. Precipitated DCU was removed by filtration, and the filtrate was evaporated in vacuo. Extractive workup [EtOAc/5% HCl(aq), 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes] and column chromatography (375 g silica gel, 1:9 EtOAc–CH_2Cl_2) yielded the mixture of dimers **12/13** (6.60 g, 66%) as a white foam: ^{1}H NMR (60 MHz, $CDCl_3$) 8.87 (d, 2 H), 6.90–7.95 (m, 29 H), 5.28 (br s, 2 H), 5.10 (br s, 8 H), 4.40–5.00 (br m, 4 H), 4.21 and 4.28 (2 s of equal intensity, 2 H), 3.52 and 3.70 (2 m of equal intensity, 2 H); IR (KBr) 3360, 3050, 1755, 1600, 1575, 1510, 1450, 1370, 1265, 1195, 1070, 965 cm^{-1}. The dimers could be separated by analytical TLC (silica gel, 1:9 EtOAc–CH_2Cl_2, R_f 0.26 and 0.23) or by preparative LC (silica gel, 1:14 EtOAc–CH_2Cl_2). Diastereomerically pure dimer samples obtained by LC showed the following: ^{1}H NMR (60 MHz, $CDCl_3$) for R_f 0.26 dimer 4.21 (s, $PhCH_2OCH_2$), 3.52 (m, $PhCH_2OCH_2$); for R_f 0.23 dimer 4.28 (s, $PhCH_2OCH_2$), 3.70 (m, $PhCH_2OCH_2$).

The mixture of dimers **12/13** could be fully deprotected by sequential treatment with Zn/HOAc and then H_2 and Pd/C, (see conditions for deprotection of monomer **10**). Deprotected dimers **14/15** show the same chromatographic mobility as naturally derived dimer[14] (cellulose TLC, 5% $NH_4^+HCOO^-$ in 0.5% HCOOH(aq), R_f for **14/15** 0.76; monomer **11** used as R_f standard).

***N*-Boc Monomer Alcohol 17.** To a suspension of MaqBr[26] (30.1 g, 100 mmol) and *N*-Boc-L-serine[25] (**16**; 23.6 g, 115 mmol) in dry THF (350 mL) at reflux was added DBU (14.9 mL, 100 mmol) over 30 min. Heating at reflux was continued for an additional 30 min, during which time the mixture became homogeneous. Cooling to ambient temperature led to crystallization of DBU·HBr; the salt was removed by filtration, and the filtrates

(44) Lindgren, B. O.; Nilsson, T. *Acta Chem. Scand.* **1973**, *27*, 888.
(45) Merz, K. W.; Fink, J. *Arch. Pharm. (Weinheim, Ger.)* **1956**, *289*, 347.

were evaporated to one-third volume. Extractive workup [EtOAc/1 M aqueous citric acid, 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes] and recrystallization from EtOAc afforded monomer **17** (39.75 g, 93.5%) as yellow crystals: mp 143–144 °C, ^{1}H NMR (60 MHz, $CDCl_3$) 8.0–8.4 (m, 4 H), 7.55–7.90 (m, 3 H), 5.60 (d, 1 H), 5.35 (s, 2 H), 4.50 (m, 1 H), 4.05 (m, 2 H), 2.87 (br t, 1 H), 1.40 (s, 9 H); IR (KBr) 3500, 3380, 2980, 1750, 1710, 1675, 1595, 1525, 1390, 1370, 1355, 1330, 1295, 1225, 1158, 1065, 932 cm^{-1}; mass spectrum, *m/e* 222 ($C_{15}H_{10}O_2^+$, MaqH$^+$), 221 ($C_{15}H_9O_2^+$, Maq$^+$), 57 ($C_4H_9^+$); $[\alpha]_D$ −15.25° (*c* 0.8, EtOH). Anal. Calcd for $C_{23}H_{23}NO_7$: C, 64.93; H, 5.45; N, 3.29. Found: C, 64.71; H, 5.49; N, 3.18. *N*-Boc monomer alcohol **17** produced as outlined above had spectral properties and an optical rotation indistinguishable from those of **17** produced from potassium salt **8** (Scheme I) by sequential reaction with MaqBr and di-*tert*-butyl dicarbonate. The procedure from *N*-Boc-L-serine, however, is more conveniently accomplished.

***N*-Boc Dimer Acid 20.** To a solution of acid monomer **18**[27] (4.87 g, 16.5 mmol), alcohol monomer **17** (6.38 g, 15.0 mmol), and HOBt (3.33 g, 21.8 mmol) in dry THF (60 mL) at 0 °C was added DCC (3.75 g, 18.2 mmol). The mixture was stirred 1 h at 0 °C and 1 h at ambient temperature. *N*-Methylmorpholine (3.62 mL, 33.0 mmol) was added and the mixture stirred 18 h at ambient temperature; TLC showed no remaining alcohol **17**. The reaction mixture was doubled in volume with EtOAc and filtered to remove DCU. Extractive workup [EtOAc/1 M aqueous citric acid, 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes] and recrystallization from 1:1 EtOAc–hexanes afforded the dimeric Maq ester **19**: 8.10 g (77%); mp 129–130 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 8.00–8.40 (m, 4 H), 7.50–7.90 (m, 3 H), 7.30 (s, 5 H), 5.50 (br s, 2 H), 5.30 (s, 2 H), 4.20–4.80 (m, 4 H), 4.47 (s, 2 H), 3.75 (d of AB q, 2 H), 1.40 (s, 18 H); IR (KBr) 3470, 3380, 2980, 1755, 1742, 1715, 1705, 1680, 1595, 1505, 1458, 1398, 1372, 1340, 1330, 1290, 1250, 1210, 1170, 1160, 935 cm^{-1}; mass spectrum, *m/e* 222 ($C_{15}H_{10}O_2$, MaqH$^+$), 221 ($C_{15}H_9O_2^+$, Maq$^+$), 91 ($C_7H_7^+$), 57 ($C_4H_9^+$).

To a solution of dimer Maq ester **19** (5.27 g, 7.50 mmol) in 3:1 EtOH–DMF (40 mL) at 0 °C were added 10% Pd on carbon (1.75 g), *N*-methylmorpholine (1.1 mL, 10 mmol), and 1,4-cyclohexadiene (5 mL, 52 mmol). The bright green suspension of catalyst was stirred overnight at ambient temperature; TLC showed no remaining Maq ester **19**. Filtration and extractive workup [EtOAc/1 M aqueous citric acid, NaCl(aq) wash] yielded an amorphous solid. Flash chromatography (CH_2Cl_2 to 5% HOAc in CH_2Cl_2 gradient) afforded dimer acid **20** (3.30 g, 91%) as a crisp white foam: ^{1}H NMR (60 MHz, $CDCl_3$) 10.45 (s, 1 H), 7.25 (s, 5 H), 5.50 (2 br overlapping d, 2 H), 4.20–4.80 (m, 4 H), 4.45 (s, 2 H), 3.65 (d of AB q, 2 H), 1.40 (s, 18 H). Dicyclohexylammonium salt: mp 141–142 °C; IR (KBr) 3380, 2970, 1745, 1720, 1705, 1640, 1580, 1500, 1390, 1370, 1340, 1250, 1170, 1060 cm^{-1}.

Coupling of *N*-Boc Dimer Acid 20 and *N*-Boc Monomer Alcohol 17. To a solution of dimer acid **20** (1.01 g, 2.10 mmol), monomer alcohol **17** (0.85 g, 2.00 mmol), and HOBt (0.424 g, 2.77 mmol) in THF (10 mL) at 0 °C was added DCC (0.476 g, 2.31 mmol). The mixture was stirred 1 h at 0 °C and 1 h at ambient temperature. *N*-Methylmorpholine (0.46 mL, 4.2 mmol) was added and the mixture stirred 18 h at ambient temperature; TLC showed only a trace of remaining alcohol **17**. Workup as for dimer **19** (vide supra) afforded an off-white foam. Column chromatography (silica gel, 1:9 EtOAc–CH_2Cl_2) of the mixture yielded three crystalline products, dehydrodimer **24** (78 mg, 7%), protected dimer **19** (493 mg, 35%), and trimer **25** (340 mg, 19%) (in order of elution). Data for **24**: mp 93–95 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 8.00–8.30 (m, 4 H), 7.50–7.85 (m, 3 H), 6.85 (br s, 1 H), 6.00 (br s, 1 H), 5.40–5.60 (m, 2 H), 5.30 (s, 2 H), 4.70 (m, 1 H), 4.55 (m, 2 H), 1.40 (s, 18 H); IR (KBr) 3380, 3430, 1750, 1720, 1675, 1595, 1510, 1325, 1295, 1165, 1065 cm^{-1}; TLC (silica gel, 1:9 EtOAc–CH_2Cl_2) R_f 0.71. Data for dimer **19**: identified by ^{1}H NMR and TLC (silica gel, 10% EtOAc in CH_2Cl_2) cospot with authentic dimer (vide supra), R_f 0.44. Data for trimer **25**: mp 159–161 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 8.00–8.40 (m, 4 H), 7.50–7.90 (m, 3 H), 7.25 (s, 5 H), 5.20–5.90 (m, 3 H), 5.30 (AB q, 2 H), 4.20–4.90 (m, 7 H), 4.55 (s, 2 H), 3.50–4.00 (m, 2 H), 1.40 (s, 27 H); IR (KBr) 3380, 2990, 1755, 1745, 1720, 1680, 1600, 1510, 1375, 1300, 1255, 1170, 1065 cm^{-1}; TLC (silica gel, 1:9 EtOAc–CH_2Cl_2) R_f 0.40. Satisfactory elemental analysis for **25** was obtained on the coupling product of dimer **30** and monomer **18** (vide infra).

***N*-Boc Monomer Acid 28.** To a solution of *N*-Boc-L-serine[25] (**16**; 3.50 g, 20.0 mmol) and *p*-bromophenacyl bromide (5.56 g, 20.0 mmol) in acetone (125 mL) was added solid $KHCO_3$ (2.0 g, 20 mmol). The suspension was stirred overnight; TLC showed only traces of phenacyl bromide remaining. Water was added to effect solution, and the mixture was evaporated in vacuo to one-quarter volume. Extractive workup [EtOAc/H_2O, 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes] afforded a white solid; trituration with boiling Et_2O yielded *N*-Boc phenacyl ester **26**: 6.65 g (85%); mp 137–138 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 7.40–7.90 (AB q, 4 H), 5.42 (br s, 1 H), 5.42 (AB q 2 H), 4.55 (m, 1 H), 4.05 (d of AB q, 2 H), 3.90 (m, 1 H, exchangeable), 1.40 (s, 9 H); IR (KBr) 3460, 3420, 2980, 1770, 1700, 1675, 1592, 1530, 1425, 1370, 1285, 1230, 1160, 1070, 970 cm^{-1}; mass spectrum, *m/e* 403 and 401 (M$^+$, ^{79}Br and ^{81}Br), 372 and 370 (M$^+$ − CH_2OH), 316 and 314 (372 and 370 − C_4H_9), 216 and 214 (*p*-BrPhCOCH$_2$OH$^+$), 57 ($C_4H_9^+$). Anal. Calcd for $C_{16}H_{20}BrNO_6$: C, 47.78; H, 5.01; N, 3.48; Br, 19.86. Found: C, 47.60; H, 5.03; N, 3.33; Br, 19.95.

To a suspension of alcohol **26** (20.1 g, 50.0 mmol) and dihydropyran (19 mL, 200 mmol) in CH_2Cl_2 (250 mL) was added pyridinium *p*-toluenesulfonate (1.20 g, 5.0 mmol). The mixture rapidly became homogeneous; after 1 h at ambient temperature, TLC showed no remaining alcohol **26**. Extractive workup [CH_2Cl_2/H_2O, NaCl(sat, aq) wash] afforded diastereomeric THP-protected monomer **27** as a syrup: ^{1}H NMR (60 MHz, $CDCl_3$) 7.50–8.00 (AB q, 4 H), 5.55 (d, 1 H), 5.35 (br s, 2 H), 4.65 (br m, 2 H), 3.50–4.20 (m, 4 H), 1.00–2.00 (m, 15 H).

Without purification, monomer **27** was dissolved in 1:19 HOAc–Et_2O (500 mL), and to the stirred solution was added zinc dust (102 g, 1.5 mol) portionwise. After 1.5 h, TLC showed no monomer **27** remaining. The suspension was filtered to remove zinc, and the filtrates were poured into H_2O (200 mL). Solid $NaHCO_3$ was added in small portions until CO_2 evolution ceased. Acid–base extractive purification [Et_2O/5% $NaHCO_3$(aq); aqueous layer acidified with $KHSO_4$ to pH 4.5 and extracted with Et_2O; organic layer washed with NaCl(sat, aq)] afforded acid **28** as a syrup. Evaporation from toluene three times removed traces of HOAc in the product, affording diastereomeric monomer acid **28** (14.81 g, 102%) as a thick syrup containing residual toluene: ^{1}H NMR (60 MHz, $CDCl_3$) 10.90 (s, 1 H), 5.60 (2 overlapping d, 1 H), 4.20–4.70 (br m, 2 H), 3.40–4.20 (m, 4 H), 1.20–2.00 (m, 15 H).

***N*-Boc Dimer Alcohol 30.** To a mechanically stirred solution of acid **28** (21.68 g, 75.0 mmol), alcohol **17** (21.25 g, 50.0 mmol), and HOBt (15.3 g, 100 mmol) in THF (200 mL) at 0 °C was added DCC (17.0 g, 82.5 mmol). The mixture was stirred 1 h at 0 °C and 1 h at ambient temperature. *N*-Methylmorpholine (16.0 mL, 150 mmol) was added and the mixture stirred 24 h; TLC showed no alcohol **17** remaining. The volume of the mixture was tripled with EtOAc and the mixture briefly heated (steam bath) to dissolve some gelatinous material. Filtration of precipitated DCU, extractive workup [EtOAc/1 M aqueous citric acid, NaCl(sat, aq), 5% $NaHCO_3$(aq), and NaCl(sat, aq) washes], and evaporation of the solvent yielded a damp solid (dimer **29**) used directly in the deprotection step. A small sample of **29** was dried in vacuo and characterized: mp 122–124 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 8.00–8.40 (m, 4 H), 7.55–7.90 (m, 3 H), 5.50 (br m, 2 H), 5.30 (s, 2 H), 3.20–4.90 (2 m, 9 H), 1.10–2.00 (m, 24 H); IR (KBr) 3480, 2940, 1755, 1740, 1720, 1710, 1680, 1595, 1570, 1430, 1300, 1160, 1060, 1030 cm^{-1}.

To the damp, crude dimer **29** (ca. 50 mmol) in 5:1 EtOH–THF (600 mL) was added pyridinium *p*-toluenesulfonate (1.25 g, 5.0 mmol), and the resulting solution was stirred overnight at 55 °C. The mixture was evaporated in vacuo to one-quarter volume and partitioned between Et_2O (500 mL) and NaCl(sat, aq) (200 mL). The Et_2O layer was washed with NaCl(sat, aq) (5 × 100 mL), dried, and evaporated to yield a yellow solid. Recrystallization from Et_2O afforded dimer **30**: 24.70 g (81% based on alcohol **17**); mp 99–100 °C; ^{1}H NMR (60 MHz, $CDCl_3$) 8.00–8.50 (m, 4 H), 7.60–8.00 (m, 3 H), 5.30–5.80 (br m, 2 H), 5.35 (s, 2 H), 4.20–4.90 (m, 4 H), 3.90 (m, 2 H), 2.60 (t, 1 H), 1.40 (s, 18 H); IR (KBr) 3400 (br), 2980, 2940, 1750, 1715, 1680, 1595, 1510, 1300, 1160, 1060 cm^{-1}. Anal. Calcd for $C_{31}H_{36}N_2O_{11}$: C, 60.78; H, 5.92; N, 4.57. Found: C, 60.52; H, 6.05; N, 4.41.

***N*-Boc Trimer Acid Alcohol 31.** To a solution of dimer alcohol **30** (24.5 g, 40.0 mmol), *O*-benzyl-*N*-(*tert*-butoxy-

carbonyl)-L-serine (18; 17.70 g, 60.0 mmol), and HOBt (12.1 g, 80.0 mmol) in dry THF (150 mL) at 0 °C was added DCC (13.60 g, 66.0 mmol). The mixture was stirred 1 h at 0 °C and 1 h at ambient temperature. *N*-Methylmorpholine (13.2 mL, 120 mmol) was added and the mixture stirred at ambient temperature for 24 h; TLC showed no remaining dimer **30**. The mixture was doubled in volume with EtOAc and precipitated DCU removed by filtration. Extractive workup as for dimer **29** (vide supra) and recrystallization from Et_2O afforded trimer **25** (23.11 g, 65%) indistinguishable (melting point, ^1H NMR, IR, TLC) from trimer produced from the coupling of dimer acid **20** and monomer alcohol **17** (vide Scheme II). Anal. Calcd for $C_{46}H_{55}N_3O_{15}$: C, 62.08; H, 6.23; N, 4.72. Found: C, 61.96; H, 6.37; N, 4.62.

A solution of protected trimer **25** (13.34 g, 15.0 mmol) in 2:6:1 THF–HOAc–H_2O (225 mL) was mixed with 10% Pd on carbon (3.50 g), and the suspension was shaken under H_2 (50 psi) overnight. The catalyst was removed by filtration, and the filtrates were evaporated in vacuo to give a brown oil. Acid–base extractive purification [EtOAc/5% $NaHCO_3$(aq); aqueous layer washed with Et_2O, acidified to pH 4.5 with $NaHSO_4$, and extracted with EtOAc; EtOAc layer washed with NaCl(sat, aq)] and recrystallization from Et_2O–hexanes afforded trimer **31** (8.68 g, 100%) in two crops: mp 113–114 °C; ^1H NMR (60 MHz, acetone-d_6) 7.75 (br s, 1 H), 6.30 (br m, 3 H), 4.55 (br m, 7 H), 3.95 (m, 2 H), 2.65 (br s, 1 H), 1.40 (s, 27 H); IR (KBr) 3350 (br), 2980, 1740, 1720, 1700, 1510, 1450, 1385, 1365, 1340, 1290, 1240, 1210, 1055 cm^{-1}. Anal. Calcd for $C_{24}H_{41}N_3O_{14}$: C, 49.74; H, 7.13; N, 7.25. Found: C, 49.50; H, 7.09; N, 6.97.

***N*-[(Benzyloxy)carbonyl] Monomer Alcohol 34.** To a refluxing solution of *N*-[(benzyloxy)carbonyl]-D-serine[28] (2.03 g, 8.49 mmol) and MaqBr (4.00 g, 13.3 mmol) in THF (35 mL) was added dropwise a solution of DBU (1.27 mL, 8.49 mmol) in THF (5 mL) over a period of 30 min. The solution was refluxed an additional 30 min and purified by extraction [$CHCl_3$/1 N HCl(aq), 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes]. Silica gel flash chromatography (CH_2Cl_2 to 1:1 EtOAc–CH_2Cl_2 gradient) of the crude product afforded monomer **34** (3.60 g, 92%) as a pale yellow solid: mp 174–175 °C (recrystallized from EtOAc); ^1H NMR (90 MHz, $CDCl_3$) 8.24 (m, 4 H), 7.74 (m, 3 H), 7.28 (s, 5 H), 5.72 (d, 1 H), 5.32 (s, 2 H), 5.10 (s, 2 H), 4.50 (m, 1 H), 4.02 (m, 2 H), 2.32 (t, 1 H, exchangeable); IR (KBr) 3520, 3290, 3060, 2950, 1725, 1690, 1670, 1590, 1545, 1345, 1325, 1290, 1260, 1215 cm^{-1}. Anal. Calcd for $C_{26}H_{21}NO_7$: C, 67.97; H, 4.61; N, 3.05. Found: C, 67.84; H, 4.69; N, 2.96.

***N*-[(Benzyloxy)carbonyl] Monomer Acid 35.** Monomer **35** was previously reported by Corey and Bhattacharyya;[24] our preparation of **35** via alcohol **34** follows. To a stirred suspension of alcohol **34** (4.71 g, 10.3 mmol) and pyridinium *p*-toluenesulfonate (262 mg, 1.04 mmol) in CH_2Cl_2 (50 mL) at ambient temperature was added dihydropyran (3.7 mL, 40.5 mmol). After 1 h the mixture had become homogeneous. The yellow solution was stirred an additional 1 h, evaporated in vacuo to a volume of ca. 50 mL, and purified by extraction [Et_2O/H_2O, NaCl(sat, aq) wash]. Evaporation of the organic layer in vacuo afforded the THP ether of monomer alcohol **34** in quantitative yield (5.60 g). The yellow solid was used in the next step without further purification.

A solution of the THP ether (2.10 g, 4.00 mmol) and *N*-methylmorpholine (2 mL, 18.1 mmol) in 2-propanol (315 mL, freeze–thaw degassed) was irradiated at 350 nm for 4 h. Evaporation of the solvent in vacuo and extractive workup [EtOAc/$NaHCO_3$(sat, aq); aqueous layer washed with EtOAc, acidified to pH 3.5 with cold 10% aqueous H_2SO_4, and extracted with CH_2Cl_2; organic layer washed with NaCl(sat, aq)] afforded diastereomeric monomer acid **35** (1.03 g, 82%) as a pale amber oil: ^1H NMR (60 MHz, $CDCl_3$) 9.9 (s, 1 H), 7.25 (s, 5 H), 5.9 (d, 1 H), 5.1 (s, 2 H), 4.3–4.8 (m, 2 H), 3.2–4.2 (m, 4 H), 1.1–1.9 (m, 6 H); IR (neat, NaCl) 3600–2300 (br), 1715, 1510, 1450, 1410, 1340, 1240, 1200 cm^{-1}.

***N*-[(Benzyloxy)carbonyl] Dimer Alcohol 36.** To a stirred solution of acid monomer **35** (0.90 g, 2.79 mmol), alcohol monomer **34** (0.843 g, 1.83 mmol), and HOBt (0.551 mg, 3.09 mmol) in THF (8 mL) at 5 °C was added DCC (0.59 mL, 3.09 mmol) dropwise. The mixture was stirred at 5 °C for 1 h and at ambient temperature for 1 h, and then *N*-methylmorpholine (0.31 mL, 2.81 mmol) was added. After being stirred 20 h, the mixture was diluted with EtOAc (20 mL) and HOAc (0.02 mL, 0.35 mmol) and stirred an additional 30 min. Filtration of precipitated DCU and extractive purification [EtOAc/1 M aqueous citric acid, $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes] yielded the dimer THP ether (1.40 g, 100%) as a pale yellow foam.

The crude dimer THP ether (1.40 g, 1.83 mmol) and pyridinium *p*-toluenesulfonate (50 mg, 0.199 mmol) were stirred at 65 °C in EtOH (20 mL) for 20 h. The solvent was evaporated to one-third volume, and the precipitated product was filtered from solution, yielding dimer alcohol **36** (1.09 g, 88%) as a pale yellow solid: mp 87–90 °C (recrystallized from EtOH); ^1H NMR (60 MHz, $CDCl_3$) 8.2 (m, 4 H), 7.7 (m, 3 H), 7.2 (s, 10 H), 6.2 (d, 1 H), 5.9 (d, 1 H), 5.25 (s, 2 H), 5.05 (s, 2 H), 5.0 (s, 2 H), 4.2–4.9 (m, 4 H), 3.9 (br s, 2 H), 3.35 (s, 1 H, exchangeable); IR ($CHCl_3$) 3420, 3020, 1740, 1715, 1670, 1590, 1500, 1450, 1320, 1290 cm^{-1}. Anal. Calcd for $C_{37}H_{32}N_2O_{11}$: C, 65.29; H, 4.74; N, 4.12. Found: C, 65.08; H, 4.68; N, 4.01.

***N*-[(Benzyloxy)carbonyl] Trimer Acid Alcohol 38.** To a stirred solution of dimer alcohol **36** (8.03 g, 11.8 mmol), monomer acid **35** (5.72 g, 17.7 mmol), and HOBt (3.62 g, 23.6 mmol) in THF (35 mL) at 5 °C was added DCC (3.72 mL, 19.5 mmol) dropwise. The mixture was stirred at 5 °C for 1 h and at ambient temperature for 1 h, *N*-methylmorpholine (3.90 mL, 35.3 mmol) was added, and stirring was continued an additional 20 h. The mixture was diluted with EtOAc (200 mL), and precipitated DCU was removed by filtration. Extractive workup [EtOAc/1 M aqueous citric acid, 5% $NaHCO_3$(aq) and NaCl(sat, aq) washes] afforded the trimer THP ether (11.63 g, 100%) as a pale yellow foam.

The crude trimer THP ether (14.69 g, 14.90 mmol) and pyridinium *p*-toluenesulfonate (0.375 g, 1.49 mmol) in 15:24 $CHCl_3$–EtOH (195 mL) were stirred at 55 °C for 23 h. The solvent was evaporated in vacuo to one-fifth volume, and the precipitated trimer alcohol **37** (12.80 g, 95%) was collected by filtration: mp 131–133 °C (recrystallized from EtOH); ^1H NMR (60 MHz, $CDCl_3$) 8.2 (m, 4 H), 7.7 (m, 3 H), 7.2 (s, 10 H), 7.15 (s, 5 H), 5.8–6.3 (m, 3 H), 5.25 (s, 2 H), 5.05 (s, 4 H), 5.0 (s, 2 H), 4.0–4.9 (m, 7 H), 3.8 (m, 2 H), 3.1 (s br, 1 H, exchangeable); IR ($CHCl_3$) 3420, 3030, 1745, 1715, 1670, 1590, 1505, 1450, 1380, 1320, 1290 cm^{-1}. Anal. Calcd for $C_{48}H_{43}N_3O_{15}$: C, 63.93; H, 4.81; N, 4.66. Found: C, 63.79; H, 4.95; N, 4.60.

A degassed (N_2 swept, 30 min) solution of trimer Maq ester **37** (3.75 g, 4.15 mmol) and *N*-methylmorpholine (2.3 mL, 21 mmol) in 23:38 $CHCl_3$–2-propanol (305 mL) was irradiated at 350 nm for 4.5 h. The yellow solution was evaporated in vacuo and purified by extraction [EtOAc/1 M citric acid(aq), NaCl(sat, aq) wash; organic layer diluted with Et_2O and washed with 5% $NaHCO_3$(aq) (10 × 60 mL); combined aqueous layers at 5 °C acidified to pH 3.5 with 10% H_2SO_4(aq) and extracted with EtOAc; organic layer washed with NaCl(sat, aq)], yielding acid alcohol **38** (1.90 g, 67%; yield of trimer in L-seryl series, 82%) as an off-white foam: ^1H NMR (250 MHz, $CDCl_3$) 7.28 (m, 15 H), 6.11–6.30 (m, 3 H), 5.25–5.85 (br s, 2 H), 5.09 (m, 6 H), 4.35–5.04 (m, 7 H), 3.95 (half an AB q, J = 11 Hz, 1 H), 3.66 (half an AB q, J = 11 Hz, 1 H); IR ($CHCl_3$) 3420, 3020, 2950, 1740, 1715, 1505, 1450, 1400, 1335, 1290, 1230, 1210, 1055 cm^{-1}; mass spectrum (field desorption), m/e 681 (M^+). The enantiomer of trimer **38** was previously reported by Corey and Bhattacharyya.[24]

***N*-[(Benzyloxy)carbonyl] Cyclic Triester 39.** The cyclization of trimer **38** follows the general procedure previously reported;[24] details of our cyclization follow. A solution of trimer **38** (0.707 g, 1.04 mmol), 4-*tert*-butyl-*N*-isopropylimidazolyl disulfide (0.454 g, 1.15 mmol), and triphenylphosphine (0.543 g, 2.07 mmol) in dry CH_2Cl_2 (8.3 mL) was stirred at ambient temperature for 15 min. The mixture was diluted with dry toluene (31 mL) and drawn into a 50-mL syringe cooled with powdered CO_2(s). The cold solution was added via syringe pump over 1.5 h to toluene (260 mL) stirred at 67 °C (internal temperature). After the addition, the solution was stirred an additional 1 h at 67 °C and evaporated in vacuo to a volume of 30 mL. The mixture was stirred with CH_3I (25 mL, passed through basic alumina) at ambient temperature for 10 min and then allowed to stand at 0 °C for 12 h. Precipitated solids were removed by filtration (Celite), and the filtrate was evaporated to a volume of 15 mL. Extractive purification [EtOAc/1 M citric acid(aq), NaCl(sat, aq), 0.2 M aqueous citrate buffer, (pH 5.5), and NaCl(sat, aq) washes] and flash chromatography (silica gel, 7:93 acetone–CH_2Cl_2) afforded

cyclic triester **39** (72 mg, 10%; yield of cyclic triester in L-seryl series, 16%) as a white foam: mp 182–183 °C (recrystallized from EtOAc–Et_2O); ^1H NMR (250 MHz, $CDCl_3$) 7.32 (s, 15 H), 6.30 (m, 1 H), 5.84 (d, 1 H), 5.10 (s, 6 H), 4.70 (m, 6 H), 4.37 (m, 3 H), 3.90 (m, 1 H); IR ($CHCl_3$) 3420, 3330, 3020, 2980, 2880, 1755, 1720, 1505, 1450, 1380, 1330, 1290, 1210, 1105, 1060 cm^{-1}; mass spectrum (field desorption), m/e 664 (M^+). Anal. Calcd for $C_{33}H_{33}N_3O_{12}$: C, 59.73; H, 5.01; N, 6.33. Found: C, 59.71; H, 5.08; N, 6.14.

Hexabenzylenantioenterobactin (41). A suspension of 10% Pd on carbon (63 mg) and cyclic triester **39** (41 mg, 0.062 mmol) in THF (4.2 mL) containing 5% (v/v) DMF was flushed with H_2. After the addition of 1.72 N HCl in dry THF (0.330 mL, 0.561 mmol), the mixture was stirred at ambient temperature under an atmosphere of H_2 for 12 h, and then the solvent was evaporated in vacuo. The residue, containing catalyst and triammonium salt **40**, was suspended in THF (3 mL), and solutions of 2,3-bis(benzyloxy)benzoyl chloride (**6**; 135 mg, 0.371 mmol) in THF (0.3 mL) and Et_3N (0.052 mL, 0.374 mmol) in THF (0.3 mL) were added simultaneously via syringe over 10 min at 0 °C. The mixture was stirred at 0 °C for 15 min and at ambient temperature for 30 min. The hydrogenation catalyst was filtered from solution (Celite), and the filtrate was purified by extraction [EtOAc/1 M citric acid(aq), 0.2 M aqueous citrate buffer (pH 5.5) and NaCl(sat, aq) washes] followed by flash chromatography (silica gel, 7:93 acetone–CH_2Cl_2), affording a mixture of hexabenzylenantioenterobactin (**41**) and 2,3-bis(benzyloxy)benzoic acid. This mixture was dissolved in CH_2Cl_2 (5 mL), and ethereal diazomethene was added until a yellow color persisted. The solution was stirred 5 min at ambient temperature and quenched with a drop of HOAc. Evaporation in vacuo and flash chromatography (silica gel, 7:93 acetone–CH_2Cl_2) afforded hexabenzylenantioenterobactin (46 mg, 61%; yield of hexabenzylenterobactin in the L-seryl series, 69%) as a white foam: ^1H NMR (250 MHz, $CDCl_3$) 8.48 (d, 3 H), 7.65 (m, 3 H), 7.08–7.45 (m, 36 H), 5.15 (half an AB q, J = 11 Hz, 3 H), 5.11 (s, 6 H), 5.04 (half an AB q, J = 11 Hz, 3 H), 4.91 (m, 3 H), 4.09 (m, 6 H); IR ($CHCl_3$) 3350, 3020, 1755, 1655, 1570, 1505, 1450, 1370, 1340, 1310, 1260, 1210 cm^{-1}. Anal. Calcd for $C_{72}H_{63}N_3O_{15}$: C, 71.45; H, 5.25; N, 3.47. Found: C, 71.61; H, 5.44; N, 3.36.

Enantioenterobactin (42). A suspension of 10% Pd on carbon (19 mg) and hexabenzylenantioenterobactin (**41**; 45 mg, 0.0372 mmol) in 2:1 THF–HOAc (2.1 mL) was shaken under H_2 (50 psi) for 20 h. The catalyst was removed by centrifugation and washed with EtOAc (5 × 3 mL). The combined supernatants were washed with 1 N HCl(aq) (4 × 4 mL) and 0.2 M aqueous citrate buffer (pH 5.5, 3 × 4 mL), dried over Na_2SO_4, and evaporated in vacuo. The residue was filtered through a short column of silicic acid (Mallinckrodt, 100-mesh; 4:1 benzene–EtOAc) to afford enantioenterobactin (17.4 mg, 70%; yield of enterobactin in the L-seryl series, 90%) as a white powder: mp 200.5–202 °C (recrystallized from EtOAc–hexanes) [lit.[5a] mp (for natural enterobactin) 202–203 °C]; ^1H NMR (250 MHz, acetone-d_6) 8.40 (d, 3 H), 7.20 (m, 3 H), 7.00 (m, 3 H), 6.72 (m, 3 H), 5.09 (m, 3 H), 4.71 (d, 6 H), 2.7–3.1 (br s, exchangeable H's); IR (KBr) 3700–2500 (br), 1745, 1635, 1580, 1530, 1455, 1340, 1260, 1170, 1070 cm^{-1}; mass spectrum (field desorption), m/e 669 (M^+), 446 (seryl dimer$^+$), 223 (seryl monomer$^+$); UV (EtOH) λ_{max} 316 (ϵ 9200). Anal. Calcd for $C_{30}H_{27}N_3O_{15}$: C, 53.82; H, 4.06; N, 6.28. Found: C, 53.50; H, 4.29; N, 6.27.

***N*-[2,3-Bis(benzyloxy)benzoyl] Monomer Acid 45.** To a stirred refluxing solution of tosyl sulfonamide **44**[32] (3.60 g, 10.05 mmol) in anhydrous ammonia (200 mL) was added sodium (1.17 g, 51.04 mmol) in small pieces. The dark blue solution was refluxed for 1 h, Bio-Rad Ag 50W-X4 (NH_4^+ form) ion-exchange resin (14.7 g) was added, and the NH_3 was allowed to evaporate overnight. The residue was dissolved in H_2O (100 mL), and the resulting solution was filtered. The filtrate was washed with $CHCl_3$ (2 × 100 mL) and the aqueous layer evaporated in vacuo to afford a white solid. To the solid and $NaHCO_3$ (2.12 g, 25.26 mmol) in H_2O (16 mL) was added dropwise 2,3-bis(benzyloxy)benzoyl chloride (**6**; 3.64 g, 10.32 mmol) in THF (5 mL). The mixture was stirred at ambient temperature for 1 h, diluted with H_2O (100 mL), and washed with Et_2O (2 × 50 mL). The aqueous layer was acidified to pH 1 with 5 N HCl(aq) and extracted with EtOAc. The organic layer was washed with NaCl(sat, aq), dried, and evaporated to yield acid monomer **45** (4.37 g, 84%) as a white foam; dicyclohexylammonium salt, mp 133–135 °C (recrystallized from EtOAc–hexanes). Data for **45**: ^1H NMR (60 MHz, $CDCl_3$) 8.70 (d, 1 H), 6.9–7.8 (s, 1 H, m, 13 H), 5.10 (m, 4 H, m, 1 H), 4.60 (m, 1 H), 3.45 (m, 2 H), 1.35 (s, 9 H); IR ($CHCl_3$) 3450, 3350, 3010, 2980, 1710, 1645, 1570, 1510, 1455, 1390, 1370, 1260, 1160 cm^{-1}.

***N*-[2,3-Bis(benzyloxy)benzoyl] Monomer Amine Salt 47.** To a stirred solution of acid monomer **45** (744 mg, 1.43 mmol) in 10:1 MeOH–H_2O (30 mL) was added a 20% (w/v) solution of Cs_2CO_3(aq) until the solution reached pH 8 (required 1.5 mL, 0.92 mmol). The solution was evaporated in vacuo and reevaporated twice from DMF (2 × 10 mL). The residue was dissolved in DMF (11 mL), *p*-bromobenzyl bromide (355 mg, 1.42 mmol) was added, and the mixture was stirred at ambient temperature for 15 h. The solvent was removed in vacuo, and the residue was triturated with H_2O and then purified by extraction [benzene/$NaHCO_3$(sat, aq), NaCl(sat, aq) wash], affording *p*-bromobenzyl ester **46** (966 mg, 98%) as a white foam: ^1H NMR (60 MHz, $CDCl_3$) 8.6 (d, 1 H), 7.0–7.7 (m, 17 H), 5.1 (s, 4 H), 5.05 (s, 2 H), 4.7 (2 m, 2 H), 3.45 (t, 2 H), 1.38 (s, 9 H).

p-Bromobenzyl ester **46** (2.86 g, 4.15 mmol) was stirred in 4.0 N HCl in THF (90 mL) for 1.5 h. Excess HCl was removed by sweeping the solution with N_2, and then the solvent was evaporated in vacuo. The residue was triturated with Et_2O and recrystallized from EtOH, yielding hydrochloride salt **47** (2.17 g, 84%) as white crystals: mp 163–166 °C; ^1H NMR (60 MHz, $CDCl_3$) 8.6 (d, 1 H), 8.0–8.6 (br s, 3 H), 6.8–7.6 (m, 17 H), 4.95 (m, 6 H), 2.9–3.8 (m, 3 H); IR (KBr) 3280, 3080, 3060, 3030, 2870, 1730, 1650, 1595, 1570, 1510, 1450, 1385, 1370, 1355, 1330, 1270, 1220 cm^{-1}.

Diastereomeric *N*-[2,3-Bis(benzyloxy)benzoyl] Dimers 48 and 49. To a stirred solution of monomer acid **45** (475 mg, 0.912 mmol), amine salt **47** (576 mg, 0.920 mmol), HOBt (251 mg, 1.857 mmol), and *N*-methylmorpholine (0.101 mL, 0.920 mmol) in THF (10 mL) at 0 °C was added DCC (0.181 mL, 0.949 mmol) dropwise. The suspension was stirred at 0 °C for 1 h and at at ambient temperature for 2 h. Precipitated DCU was filtered from the solution and the filtrate evaporated in vacuo. Extractive workup [EtOAc/1 N HCl(aq), $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes] afforded a white form. The remaining DCU was removed by dissolving the dimers in EtOAc and filtering the precipitate. Evaporation of the solvent yielded the dimers **48**/**49** (1.036 g, 100%) as a white foam: ^1H NMR (60 MHz, $CDCl_3$) 8.5 (m, 2 H), 6.9–7.7 (m, 31 H), 5.05 (s, 8 H), 5.0 (s, 2 H), 4.75 (m, 1 H), 4.2–4.6 (m, 2 H), 3.1–3.8 (m, 4 H), 1.35 (s, 9 H); IR (KBr) 3330, 3080, 3060, 3030, 2970, 2910, 1735, 1705, 1660, 1640, 1575, 1520, 1450, 1430, 1375, 1310, 1260, 1210, 1190 cm^{-1}. TLC (silica gel, 1:1 hexanes–EtOAc) showed the dimers as two closely running spots, R_f 0.21 and 0.25; analytical LC (μ-Porasil, 3:1 CH_2Cl_2–EtOAc) showed two peaks of equal area. The dimer mixture was indistinguishable from authentic diastereomeric dimer made from racemic acid (cf. **45**) and chirally pure monomer **47**. Fractional recrystallization of the mixture **48**/**49** gave two crops (mp 153–159 and 65–70 °C, respectively). The higher melting crop showed a 9:1 enrichment in the slower eluting peak (LC); the lower melting crop showed a 9:1 enrichment in the faster eluting peak (LC). The ^1H NMR (60 MHz, $CDCl_3$) of each crop was indistinguishable from the ^1H NMR of the crude 1:1 mixture of **48** and **49**.

Monomer Amine Salt 52. To a refluxing solution of monomer acid **50** (2.57 g, 7.60 mmol) and MaqBr (3.66 g, 12.2 mmol) in THF (45 mL) was added dropwise a solution of DBU (1.14 mL, 7.62 mmol) in THF (5 mL) over 30 min. The mixture was refluxed an additional 30 min, cooled to ambient temperature, and purified by extraction [$CHCl_3$/1 N HCl(aq), $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes]. The product was purified further by silica gel flash chromatography (CH_2Cl_2 to 3:1 CH_2Cl_2–EtOAc gradient), affording fully protected monomer **51** (3.70 g, 87%) as a pale yellow solid: mp 176.5–177.5 °C (recrystallized from EtOAc); ^1H NMR (60 MHz, $CDCl_3$) 8.3 (m, 4 H), 7.8 (m, 3 H), 7.3 (s, 5 H), 6.1 (d, 1 H), 5.4 (s, 2 H), 5.2 (m, 1 H), 5.15 (s, 2 H), 4.6 (m, 1 H), 3.6 (m, 2 H), 1.4 (s, 9 H); IR (KBr) 3350, 2980, 2960, 1740, 1670, 1590, 1520, 1440, 1420, 1385, 1360, 1350, 1290, 1260, 1035 cm^{-1}.

A solution of monomer **51** (3.60 g, 6.45 mmol) in 7:3 CF_3CO-OH–H_2O (30 mL) was stirred at room temperature for 3.5 h and then evaporated in vacuo. Reevaporation from toluene (three times) and trituration with Et_2O afforded monomer amine salt **52** (3.20 g, 87%) as a pale yellow powder: mp 169–170.5 °C

(recrystallized from EtOH); ^{1}H NMR (60 MHz, Me_2SO-d_6) 8.4–8.7 (br s, 4 H), 7.6–8.4 (m, 7 H), 7.4 (s, 5 H), 5.4 (s, 2 H), 5.2 (s, 2 H), 4.8 (m, 1 H), 3.4 (m, 2 H); IR (KBr) 3380, 3030, 2920, 1735, 1720, 1660, 1585, 1520, 1450, 1425, 1320, 1290, 1200 cm^{-1}. Anal. Calcd for $C_{28}H_{23}N_2O_8F_3$: C, 58.74; H, 4.05; F, 9.96; N, 4.89. Found: C, 58.44; H, 4.27; F, 10.10; N, 5.08.

Dimer Amine Salt 54. To a solution of acid monomer **50** (1.92 g, 5.67 mmol) and HOBt (1.07 g, 6.97 mmol) in DMF (8 mL) at 0 °C was added DCC (1.11 mL, 5.82 mmol) dropwise. The mixture was stirred at 0 °C for 1 h and at ambient temperature for 1 h, and then a solution of amine salt **52** (3.20 g, 5.58 mmol) and *N*-methylmorpholine (0.62 mL, 5.62 mmol) in DMF (8 mL) was added. After being stirred 20 h, the mixture was warmed to dissolve precipitated dimer; DCU was filtered from the warm solution and washed with DMF. The filtrate was evaporated to one-tenth volume and purified by extraction [EtOAc/1 M aqueous citric acid, $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes], yielding dimer **53** as a pale yellow solid in quantitative yield (4.50 g).

Crude dimer **53** (4.50 g, 5.58 mmol) was dissolved in 7:3 CF_3COOH–H_2O (40 mL) and stirred at ambient temperature for 2.5 h. Evaporation of the solvent and reevaporation of the residue from toluene (three times), trituration with Et_2O, and recrystallization from EtOH afforded dimer amine salt **54** (3.84 g, 87%) as a pale yellow solid: mp 157–161 °C; ^{1}H NMR (60 MHz, $CDCl_3$ + Me_2SO-d_6) 8.3 (m, 4 H), 7.9 (m, 3 H), 7.4 (s, 10 H), 5.4 (s, 2 H), 5.2 (s, 4 H), 4.5 (m, 2 H), 3.7 (m, 2 H), 3.3 (m, 2 H); IR (KBr) 3340, 3080, 3040, 2960, 1750, 1720–1660, 1610, 1595, 1535, 1460, 1330, 1300, 1210, 1055, 935 cm^{-1}. Anal. Calcd for $C_{39}H_{35}N_4O_{11}F_3$: C, 59.09; H, 4.45; N, 7.07; F, 7.19. Found: C, 59.38; H, 4.49; N, 7.06; F, 6.81.

Trimer Amino Acid Salt 57. To a solution of monomer acid **50** (1.65 g, 4.89 mmol) and HOBt (925 mg, 6.05 mmol) in DMF (8 mL) at 0 °C was added DCC (0.96 mL, 5.03 mmol) dropwise. The mixture was stirred at 0 °C for 1 h and at ambient temperature for 1 h, and a solution of amine salt **54** (3.83 g, 4.83 mmol) and *N*-methylmorpholine (0.54 mL, 4.89 mmol) in DMF (8 mL) was added. The mixture was stirred 20 h, warmed to dissolve precipitated trimer, and filtered to remove precipitated DCU. The filtrate was evaporated in vacuo and the residue purified by extraction [$CHCl_3$/1 M aqueous citric acid, $NaHCO_3$(sat, aq) and NaCl(sat,aq) aq) washes], yielding trimer **55** (4.29 g, 89%) as a pale yellow solid.

Crude trimer **55** (5.38 g, 5.39 mmol) in freshly distilled, degassed 1,4-dioxane (300 mL) was treated with a solution of $Na_2S_2O_4$ (12.2 g, 70.1 mmol) in 5% $NaHCO_3$ (150 mL, pH of solution 7.2). The mixture was stirred at ambient temperature for 1 h, H_2O (100 mL) was added, and stirring was continued an additional hour. Dioxane was removed in vacuo, and the resulting aqueous mixture was extracted with EtOAc [1 M aqueous citric acid, H_2O and NaCl(sat, aq) washes]. The residue from evaporation of the organic layer was purified by silica gel column chromatography (5% HOAc in CH_2Cl_2 to 5% HOAc in 3:1 EtOAc–CH_2Cl_2 gradient), affording trimer acid **56** (4.03 g, 95%) as a white powder: mp 158–160 °C (recrystallized from EtOH–Et_2O); ^{1}H NMR (60 MHz, CD_3OD + Me_2SO-d_6) 7.4 (s, 15 H), 5.2 (s, 6 H), 4.55 (exchangeable H's), 4.1–4.5 (m, 3 H), 3.3–3.7 (m, 6 H), 1.45 (s, 9 H); IR (KBr) 3700–2750 (br), 1720–1650, 1525, 1455, 1440, 1400, 1370, 1350, 1260, 1170, 1060 cm^{-1}. Anal. Calcd for $C_{38}H_{46}N_6O_{12}$: C, 58.60; H, 5.95; N, 10.79; O, 24.65. Found: C, 58.58; H, 5.96; N, 10.52; O, 24.40.

To a stirred solution of 2.1 M HCl in EtOAc (35 mL) was added finely powdered trimer acid **56** (2.23 g, 2.86 mmol). The mixture was diluted with EtOAc (15 mL), stirred at room temperature for 1 h, diluted with dry Et_2O (150 mL), and centrifuged. The pellet was washed with Et_2O (5 × 25 mL) and dried to afford amino acid salt **57** (1.80 g, 88%) as a white powder: mp 211.5–213 °C (recrystallized from MeOH/Et_2O); ^{1}H NMR (60 MHz, CD_3OD) 7.2 (s, 15 H), 5.05 (s, 2 H), 5.0 (s, 4 H), 4.7 (exchangeable H's), 4.1–4.6 (m, 3 H), 3.2–3.8 (m, 6 H); IR (KBr) 3700–2300, 3300, 3030, 2940, 1720, 1690, 1660, 1520, 1445, 1385, 1290, 1260, 1230, 1180, 1020 cm^{-1}. Anal. Calcd for $C_{33}H_{39}N_6O_{10}Cl$: C, 55.42; H, 5.50; N, 11.75; Cl, 4.96. Found: C, 55.20; H, 5.40; N, 11.52; Cl, 5.23.

Cyclo(tri-β-alanine) (60). (A) Diphenylphosphoryl Azide Closure. A solution of tri-β-alanine hydrochloride (**59**, 150 mg, 0.561 mmol) in DMF (110 mL) was evaporated to 80 mL in vacuo and cooled to –40 °C, and diphenylphosphoryl azide (145 μL, 0.673 mmol) was added via syringe. The solution was stirred at –40 °C, and Et_3N was added until the pH, measured with premoistened, narrow-range pH paper, was greater than 7.0. The mixture was allowed to stand at –25 °C for 2 days and at 0 °C for 4 days. The pH was maintained above 7.0 during this time by the addition of Et_3N every 12 h. The mixture was diluted with H_2O (10 mL), stirred with Bio-Rad RG 501-X8 (20–50 mesh) mixed-bed resin for 24 h, filtered, evaporated in vacuo, and recrystallized from H_2O (1 mL) to afford cyclic trimer **60** (17.1 mg, 14.3%) as colorless needles: mp >360 °C; ^{1}H NMR (60 MHz, D_2O) 3.5 (t, 6 H), 2.5 (t, 6 H); IR (KBr) 3300, 3100, 2990, 2940, 2880, 2850, 2480, 2440, 1660, 1565, 1445, 1430, 1370, 1345, 1285, 1245, 1200 cm^{-1}; mass spectrum, *m/e* 213 (M^+), 185 (M^+ – CO or M^+ – C_2H_4).

(B) DCC/HOBt Closure. To a solution of tri-β-alanine hydrochloride (**59**; 489 mg, 1.83 mmol) at 0 °C were added HOBt (327 mg, 2.14 mmol), *N*-methylmorpholine (200 μL, 1.81 mmol), and DCC (0.38 mL, 1.99 mmol). The mixture was allowed to warm to ambient temperature and was stirred for 110 h. Precipitated polymer was filtered from solution and the filtrate evaporated in vacuo, affording a white powder. Recrystallization from H_2O yielded cyclic trimer **60** (32 mg, 8%) as colorless needles: mp >360 °C; other data, vide supra.

Fully Protected *N*-[2,3-Bis(benzyloxy)benzoyl] Trimer 63. To a solution of trimer acid **56** (0.38 g, 0.49 mmol) in 9:1 CH_2Cl_2–MeOH (20 mL) was added ethereal diazomethane until a yellow color persisted. After the mixture was stirred 10 min, excess CH_2N_2 was quenched with a few drops of HOAc and the mixture evaporated in vacuo. Silica gel column chromatography (1:1 EtOAc–CH_2Cl_2) yielded trimer methyl ester **61** (340 mg, 88%) as a white solid: mp 155–157 °C; ^{1}H NMR (250 MHz, $CDCl_3$) 7.32 (m, 15 H), 7.13 (m, 1 H), 7.01 (m, 1 H), 6.75 (m, 1 H), 6.21 (m, 2 H), 5.12 (m, 6 H, m, 1 H), 4.47 (m, 1 H), 4.26 (m, 2 H), 3.98 (m, 1 H), 3.78 (s, 3 H, m, 2 H), 3.53 (m, 2 H), 3.30 (m, 1 H), 1.35 (s, 9 H); IR (KBr) 3300, 3080, 3060, 3030, 2970, 2950, 1740–1640, 1520, 1450, 1430, 1385, 1360, 1340, 1245, 1160, 1040 cm^{-1}.

A suspension of trimer methyl ester **61** (200 mg, 0.252 mmol) and 10% Pd on carbon (140 mg) in 3:2 THF–HOAc (50 mL) was flushed with H_2 and shaken at 40 psi of H_2 for 18 h. The catalyst was filtered from solution, and the filtrate was evaporated in vacuo, stripped from toluene (three times) in vacuo, and triturated with EtOAc to afford 130 mg of triammonium acetate **62** (91%) as a white powder: ^{1}H NMR (60 MHz, D_2O + CD_3CN) 4.4 (exchangeable H's and methine protons), 3.2–4.0 (m, 6 H, s, 3 H), 1.95 (s, 9 H), 1.45 (s, 9 H).

To a stirred suspension of triammonium acetate **62** (190 mg, 0.333 mmol) in THF (5 mL) at 0 °C were added simultaneously solutions of 2,3-bis(benzyloxy)benzoyl chloride (**6**; 788 mg, 2.14 mmol) in THF (2 mL) and Et_3N (0.290 mL, 2.09 mmol) in THF (2 mL) over a period of 20 min. The resulting suspension was stirred at 0 °C for 10 min and at room temperature for 1 h. Extractive workup [EtOAc/1 M aqueous citric acid, $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes] and silica gel flash chromatography yielded a mixture of trimer **63** and 2,3-dihydroxybenzoic acid. This mixture was dissolved in CH_2Cl_2 (10 mL), treated with ethereal diazomethane until a yellow color persisted for 10 min, quenched with HOAc, evaporated under reduced pressure, and purified by silica gel flash chromatography (CH_2Cl_2 to 5% EtOH in CH_2Cl_2 gradient) to afford pure trimer **63** (298 mg, 67%) as a white solid: mp 183.5–186 °C (recrystallized from CH_2Cl_2–Et_2O); ^{1}H NMR (250 MHz, $CDCl_3$) 8.82 (d, 1 H), 8.58 (t, 2 H), 6.98–7.77 (m, 41 H), 5.13 (m, 13 H), 4.88 (m, 1 H), 4.47 (m, 1 H), 4.28 (m, 1 H), 3.84 (m, 1 H), 3.66 (s, 3 H), 3.09 (m, 5 H), 1.40 (s, 9 H); IR (KBr) 3330, 3100, 3080, 3040, 2940, 2880, 1745, 1710, 1685, 1660, 1655, 1580, 1520, 1455, 1375, 1310, 1265, 1210, 1170, 1130, 1090, 1030 cm^{-1}. Anal. Calcd for $C_{78}H_{78}N_6O_{15}$: C, 69.94; H, 5.87; N, 6.27. Found: C, 69.92; H, 6.03; N, 6.26.

Linear Enterobactin Analogue 66. A solution of trimer **63** (230 mg, 0.172 mmol) in dry formic acid (5 mL) was stirred at room temperature for 2 h, evaporated in vacuo, and triturated with anhydrous Et_2O to afford formate salt **64** (201 mg, 90.9%) as a white foam: ^{1}H NMR (250 MHz, $CDCl_3$) 8.75 (t, 2 H), 8.63 (d, 1 H), 6.98–7.70 (m, 41 H), 5.12 (m, 12 H), 4.82 (m, 1 H), 4.52 (m, 1 H), 4.17 (m, 1 H), 3.69 (m, 1 H), 3.66 (s, 3 H), 3.42 (m, 1 H), 3.32 (m, 2 H), 2.72 (m, 1 H), 2.64 (m, 1 H); IR ($CHCl_3$) 3340, 3060, 3000, 2940, 2870, 1730, 1715, 1680, 1650, 1570, 1515, 1495,

1450, 1370, 1310, 1260, 1210, 1140, 1080, 1040, 1025 cm^{-1}.

Formate salt **64** (51 mg, 0.040 mmol) in pyridine (1 mL) and acetic anhydride (1 mL) was stirred at room temperature for 2 h. Purification by extraction [EtOAc/1 N HCl(aq), $NaHCO_3$(sat, aq) and NaCl(sat, aq) washes] and silica gel column chromatography (7:93 EtOH-CH_2Cl_2) afforded *N*-acetyl trimer **65** (37 mg, 72%) as a white solid: ^1H NMR (250 MHz, $CDCl_3$) 8.70 (d, 1 H), 8.49 (d, 2 H), 6.9-7.7 (m, 41 H), 6.11 (t, 1 H), 5.10 (m, 12 H), 4.79 (m, 1 H), 4.44 (m, 1 H), 4.31 (m, 1 H), 3.63 (s, 3 H), 3.2-3.6 (m, 6 H), 1.47 (s, 3 H); IR ($CHCl_3$) 3340, 3060, 3030, 2930, 2870, 1735, 1650, 1570, 1510, 1450, 1370, 1310, 1260, 1210 cm^{-1}.

A suspension of trimer **65** (37 mg, 0.0289 mmol) and 10% Pd on carbon (8 mg) in 1:1 CH_2Cl_2-MeOH (4 mL) was flushed with hydrogen, and 2.38 N HCl in MeOH (18 μL, 0.043 mmol) was added. The mixture was stirred at room temperature under an atmosphere of hydrogen for 12 h, and the catalyst was filtered (Celite) and washed with EtOAc. The combined filtrates were washed with 1 N HCl(aq) and 0.2 M aqueous citrate buffer (pH 5.5). The organic phase was dried (Na_2SO_4), evaporated under reduced pressure, and recrystallized from EtOAc/hexane to afford linear trimer **66** (16 mg, 75%) as a white powder: mp 155-157 °C (precipitated from MeOH with H_2O and lyophilized); ^1H NMR (250 MHz, CD_3OD) 7.18-7.27 (m, 3 H), 6.88-6.94 (m, 3 H), 6.64-6.74 (m, 3 H), 4.75 (m, 1 H), 4.60 (m, 2 H), 3.86 (m, 1 H), 3.72 (s, 3 H), 3.50-3.65 (m, 5 H), 1.87 (s, 3 H); IR (KBr) 3700-2500 (br), 1730, 1635, 1580, 1540, 1525, 1450, 1360, 1330, 1260, 1170 cm^{-1}; mass spectrum (field desorption), *m/e* 740 (M^+).

Δ-Cis Fe(III) Complex 67. Linear trimer **66** (0.9 mg, 1.22 μmol) in MeOH (3.0 mL) was mixed with 4×10^{-4} M $FeCl_3$ in MeOH (3.0 mL). The resulting violet solution was diluted 1:1 with 0.1 M aqueous phosphate buffer (pH 7.2) to afford a wine-red solution of ferric complex **67**: visible spectrum (3:1 pH 7.2 aqueous phosphate buffer-MeOH) λ_{max} 495 nm (ε 4400); CD spectrum, vide Figure 1.

Equal volumes of ferric complex **67** (1×10^{-4} M) in 50% aqueous MeOH and ethylenediaminetetraacetic acid (EDTA) disodium salt in 0.1 M aqueous phosphate buffer (pH 7.2) were mixed, and the absorption at 495 nm was measured after equilibration of chelated Fe(III) was observed (constant optical density at 495 nm). The absorbance at 495 nm was 4%, 9%, and 86% diminished at 10^{-3}, 10^{-2}, and 10^{-1} M EDTA, respectively. By use of these data and a pK_a of 10.1 ± 0.2 for linear analogue **66**, the formation constant was calculated; $K_f = 10^{46.5 \pm 1.2}$ (K_f for Fe(III)·EDTA complex = 10^{25}).[46]

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Registry No. **5**, 4726-96-9; **6**, 69146-58-3; **7**, 78088-92-3; **8**, 40351-09-5; **9**, 78088-93-4; **10**, 78088-94-5; **11**, 7724-78-9; **12**, 78088-95-6; **13**, 78148-29-5; **14**, 30414-15-4; **15**, 78148-30-8; **16**, 3262-72-4; **17**, 78088-96-7; **18**, 23680-31-1; **19**, 78088-97-8; **20**, 78088-98-9; **20** dicyclohexylammonium salt, 78088-99-0; **24**, 78089-00-6; **25**, 78089-01-7; **26**, 78089-02-8; **27** (isomer 1), 78089-03-9; **27** (isomer 2), 78089-04-0; **28** (isomer 1), 78089-05-1; **28** (isomer 2), 78089-06-2; **29**, 78089-07-3; **30**, 78089-08-4; **31**, 78089-09-5; **33**, 6081-61-4; **34**, 75299-15-9; **34** THP ether, 78089-10-8; **3** (isomer 1), 78148-31-9; **35** (isomer 2), 78148-32-0; **36**, 75299-17-1; **36** THP ether, 78089-11-9; **37**, 75299-18-2; **37** THP ether, 78089-12-0; **38**, 75299-19-3; **39**, 75363-09-6; **40**, 75363-10-9; **41**, 75299-20-6; **42**, 75363-11-0; **44**, 16947-86-7; **45**, 78089-13-1; **45** dicyclohexylammonium salt, 78089-14-2; **46**, 78089-15-3; **47**, 78089-16-4; **48**, 78089-17-5; **49**, 78148-33-1; **50**, 16947-84-5; **51**, 78089-18-6; **52**, 78089-20-0; **53**, 78089-21-1; **54**, 78089-23-3; **55**, 78089-24-4; **56**, 78089-25-5; **57**, 78089-26-6; **59**, 78089-27-7; **60**, 10491-78-8; **61**, 78089-28-8; **62**, 78089-30-2; **63**, 78089-31-3; **64**, 78089-33-5; **65**, 78089-34-6; **66**, 78089-35-7; **67**, 78090-01-4; 2,3-dihydroxybenzaldehyde, 24677-78-9; benzyl chloride, 100-44-7; 2,3-bis(benzyloxy)benzaldehyde, 5779-91-9; 2,3-bis(benzyloxy)benzoic acid, 74272-78-9; oxalyl chloride, 79-37-8; *p*-bromophenacyl bromide, 99-73-0; 2-(bromomethyl)anthraquinone, 7598-10-9; *p*-bromobenzyl bromide, 589-15-1.

(46) O'Brien, I. G.; Cox, G. B.; Gibson, F. *Biochim. Biophys. Acta* 1971, *237*, 537.

Convenient Synthesis of the Pseudoguaiane Ring System

J. Joseph Christie, Thankamma E. Varkey, and John A. Whittle*

Department of Chemistry, Lamar University, Beaumont, Texas 77710

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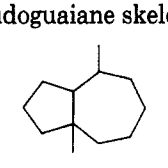
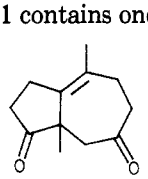
Ketone 8 was conveniently synthesized from acyclic precursors in a four-step procedure. Condensation of *cis*-1,4-dichloro-2-butene with diethyl ketone gave 2,7-dimethyl-4-cycloheptenone, which was alkylated with propargyl bromide. The triple bond was hydrated with mercuric ion impregnated Dowex 50 and the compound cyclized with potassium *tert*-butoxide to yield ketone 8.

The pseudoguaiane skeleton 1 contains one of the more



1



2

common ring systems found in hydroazulenic sesquiterpenoids.[1] Many early syntheses of the hydroazulenic ring system involved "special" reactions such as rearrangements, ring expansions and photolyses. Over the past decade, however, many syntheses using "conventional" procedures have appeared.[2] Recently, Lansbury[3] has used the different reactivities of the carbonyls in diketone 2,

(1) See: Devon, T. K; Scott, A. I. "Handbook of Naturally Occurring Compounds"; Academic Press: New York, 1972; Vol. II, pp 61-72.

(2) See, for example: Marshall, J. A.; Partridge, J. J. *Tetrahedron* 1969, *25*, 2159. Kato, M.; Kosugi, H.; Yoshikoshi, A. *Chem. Commun.* 1970, 185. Hendrickson, J. B.; Boeckman, R. K., Jr. *J. Am. Chem. Soc.* 1971, *93*, 1307. Heathcock, C. H.; Ratcliffe, R.; Van, J. *J. Org. Chem.* 1972, *37*, 1976. Marshall, J. A.; Ruden, R. A. *Ibid.* 1971, *36*, 2569. Marshall, J. A.; Green, A. E. *Ibid.* 1972, *37*, 982. Anderson, N. H.; Uh, H.-S. *Synthe. Commun.* 1973, *3*, 115. Anderson, N. H.; Uh, H.-S. *Tetradron Lett.* 1973, 2079. Buchanan, G. L.; Young, G. A. R. *J. Chem. Soc., Perkin Trans. 1*, 1974, 2404. Marshall, J. A.; Ruth, J. A. *J. Org. Chem.* 1974, *39*, 1971. De Clercq, P.; Vandewalle, M. *Ibid.* 1977, *42*, 3447.

(3) Lansbury, P. T.; Serelis, A. K. *Tetrahedron Lett.* 1978, 1909.

0022-3263/81/1946-3590$01.25/0