Total Synthesis of (+)-Duocarmycin A, *epi*-(+)-Duocarmycin A and Their Unnatural Enantiomers: Assessment of Chemical and Biological Properties

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Abstract: Full details of an enantioselective total synthesis of (+)-duocarmycin A (**1**) are described in which a solution to the control of the relative and absolute stereochemistry of the remote stereocenters is provided. Catalytic asymmetric dihydroxylation of **15** was employed to introduce the absolute stereochemistry required for the activated cyclopropane, and a diastereoselective Dieckmann-type condensation of **61** was employed to control the absolute stereochemistry of the C6 quaternary center. The complementary diastereoselectivity of a thermodynamic versus kinetic condensation of **61** permitted the divergent synthesis of (+)-duocarmycin A or *epi*-(+)-duocarmycin A from common intermediates. Final introduction of the reactive cyclopropane was accomplished by transannular spirocyclization of the mesylate **44** upon treatment with base or directly from the corresponding free alcohol itself, duocarmycin D_1 (**42**), upon Mitsunobu activation. Notably, the asymmetric dihydroxylation of **15** employing $(DHQD)_2$-PHAL/$(DHQ)_2$-PHAL was found to proceed with a reverse enantioselectivity than predicted from established models. Employing this approach, the key partial structures (+)-*N*-BOC-DA (**67**) and (+)-6-*epi*-*N*-BOC-DA (**71**) and their unnatural enantiomers were also prepared, and a study of their acid-catalyzed solvolysis reactivity, regioselectivity (3:2), and stereochemistry is detailed. Notably, the solvolysis reaction regioselectivity is lower than the characteristic adenine N3 alkylation of duplex DNA, which proceeds with exclusive nucleophilic addition to the least substituted C8 cyclopropane carbon. This may be attributed to the significant destabilizing torsional strain and steric interactions characteristic of the S_N2 reaction of a large nucleophile that accompany the abnormal addition of adenine when restricted to the minor groove bound orientation of the reactants.

Two independent reports detailed the isolation and structure determination of the initial members of a new class of exceptionally potent antitumor antibiotics now including duocarmycin A (**1**), its potent ring open derivatives **2**−**5**, and duocarmycin SA (**6**), Figure 1.[1−5] The single-crystal X-ray structure determination[4] of duocarmycin C_2 (**4**, pyrindamycin A) established the relative and absolute configuration of the remote stereocenters, which through chemical interconversions[1,10] provided the absolute stereochemistry for **1**−**5**. In the short time since their disclosure, they have been shown to exert their biological effects[1−4,6−8] through a characteristic sequence-selective DNA alkylation.[5,9−17]

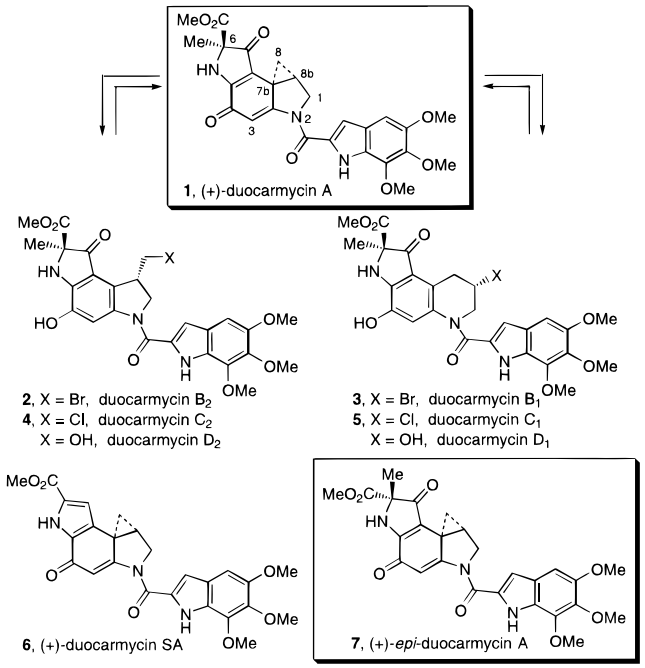

Figure 1.

Duocarmycin A (**1**) constitutes the most reactive and challenging member of this class of agents, and approaches to the

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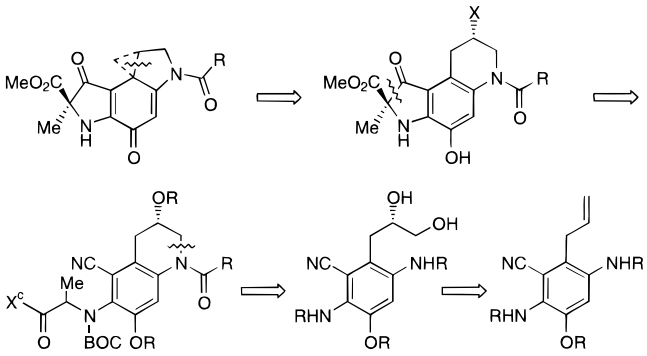
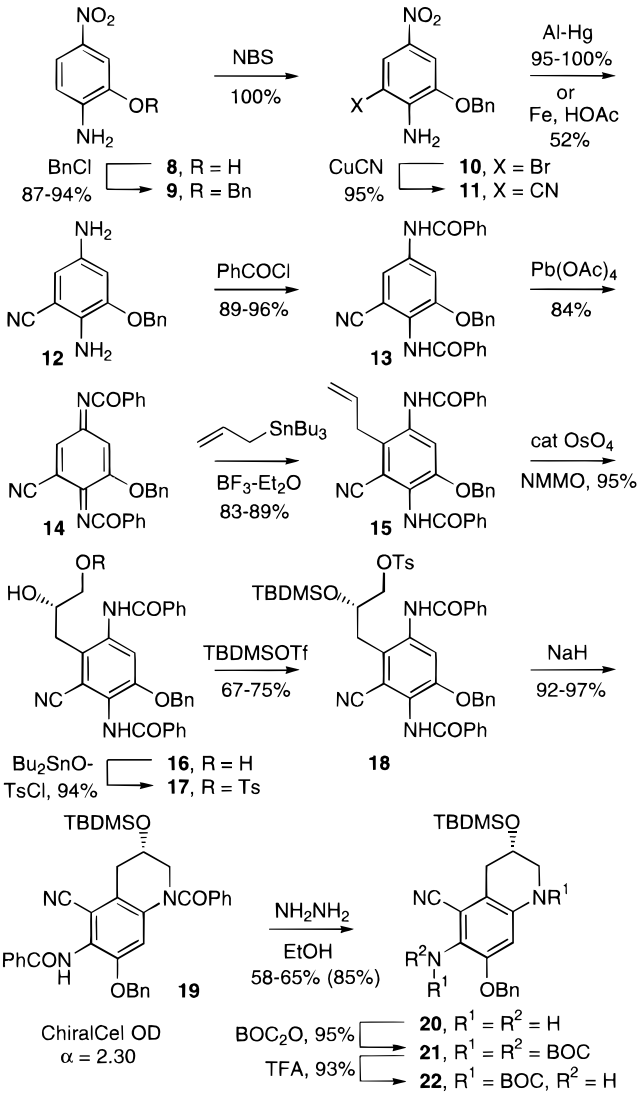
control of the relative and absolute stereochemistry of its remote stereocenters have not been forthcoming. Herein, we report full details of an enantioselective total synthesis of (+)-duocarmycin A (**1**) and *epi*-(+)-duocarmycin A (**7**) and their unnatural enantiomers in which a solution to the control of the remote stereochemistry is provided.[18] The efforts proceeded through duocarmycin D_1 as a key intermediate and complement the nondiastereoselective synthesis of **1** and **7** described by Terashima and co-workers[19] as well as related efforts on duocarmycin SA.[20,21]

Central to the strategy is the ability to prepare both enantiomers of the cyclopropane stereochemistry while maintaining the opportunity to independently control the C6 absolute stereochemistry, thereby providing access to the natural and unnatural enantiomers of either **1** or **7**. Key elements of the approach include a diastereoselective Dieckmann-like condensation for introduction of the C6 quaternary center and a transannular Ar-3' spirocyclization[1,10,12,21,22] of duocarmycin D_1 for introduction of the reactive cyclopropane, a process that has also been reported in the context of CC-1065 analog synthesis.[23] In turn, the absolute stereochemistry of the cyclopropane was set through use of an asymmetric dihydroxylation reaction[23] that was found to proceed with an enantioselectivity opposite that predicted from established models (Scheme 1). Employing Evans' optically active acyloxazolidinones, a Dieckmann-like reaction permitted the introduction of either the natural 6(*R*) or *epi*-6(*S*) stereochemistry through use of the enantiomeric auxiliaries or, more interestingly, by conducting the reaction of a given auxiliary under thermodynamic versus kinetic reaction conditions for this reversible reaction.

Scheme 1



Scheme 2



Nondiastereoselective Total Synthesis of Duocarmycin A and *epi*-Duocarmycin A. Prior to initiating our efforts on an asymmetric synthesis of **1** and **7**, we first examined the basic elements of the approach with the nondiastereoselective preparation of duocarmycin A (**1**) and *epi*-duocarmycin A (**7**). Conversion of commercially available 2-amino-5-nitrophenol to **13**[24,25] preceeded oxidation to the key *p*-quinodiimide **14** (84%) effected by $Pb(OAc)_4$ (Scheme 2). Regiospecfic Lewis

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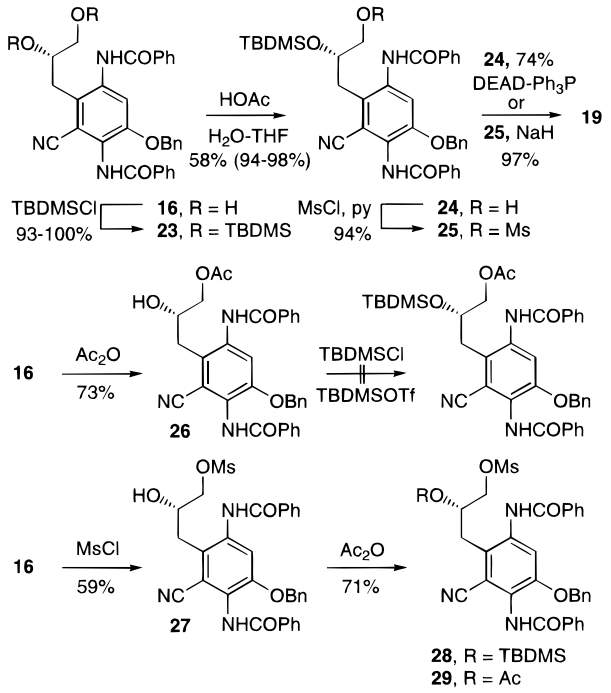
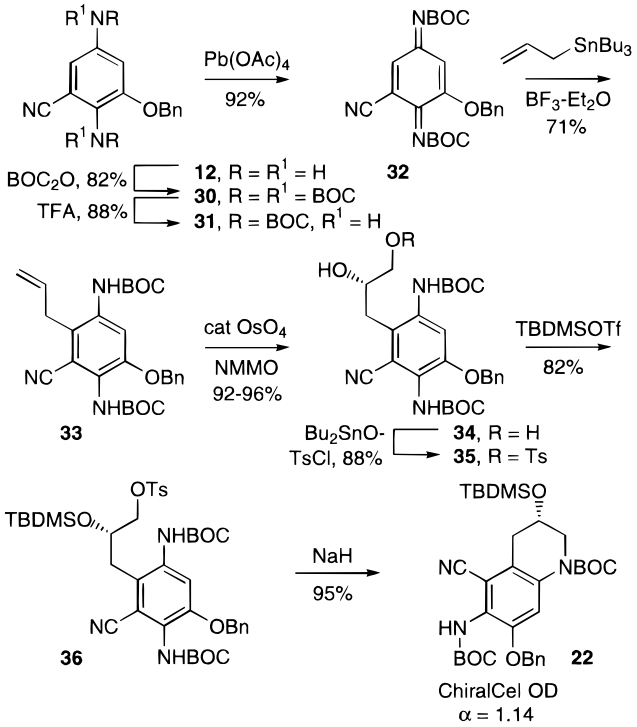
(25) Full experimental details and characterization are provided in the Supporting Information.

Scheme 3



Scheme 4



acid-catalyzed[24] addition of allyltributyltin (0.5 equiv of $BF_3{\cdot}OEt_2$, CH_2Cl_2, −20 °C, 3 h, 83−89%) cleanly provided **15**. Catalytic dihydroxylation of **15** through treatment with OsO_4 (0.2 equiv) in the presence of *N*-methylmorpholine *N*-oxide (NMMO, 2 equiv) in aqueous acetone provided diol **16** (95%). Selective formation of the primary tosylate **17** (1.1 equiv of Bu_2SnO, toluene−THF 10:1, reflux, 6 h; 1.2 equiv of TsCl, cat. Et_3N, 25 °C, 12 h, 87−94%) followed by subsequent protection of the secondary alcohol as its TBDMS ether (67−75%) cleanly provided **18**. Cyclization of **18** to afford the first key intermediate **19** was accomplished by base-promoted closure (2 equiv of NaH, THF, 0 °C, 2 h, 92−97%). Deprotection of the two benzoyl protecting groups (NH_2NH_2−EtOH 10:1, 58−65%) followed by immediate reprotection of the free amine **20** with BOC_2O (95%) provided **21**. The major byproduct of the benzoyl deprotection reaction was the free alcohol of the amine **20** resulting from additional desilylation under the vigorous reaction conditions. Simply subjecting this recovered alcohol to TBDMSOTf treatment (2,6-lutidine, CH_2Cl_2) followed by a brief treatment with 1 M aqueous citric acid afforded additional **20** and overall conversions of 85% for the transformation of **19** to **21**. Mild acid-catalyzed removal of the labile-BOC group of **21** (5−6 equiv TFA, CH_2Cl_2, 93%) cleanly provided **22**. Less vigorous conditions for acylation of the amine **20** led to mono BOC formation at the more reactive secondary amine with no evidence for the generation of **22** and the more vigorous conditions employing DMAP preferentially provided **21**.

The key intermediate **19** was also obtained by exhausive protection of the diol **16** as its bis TBDMS ether **23**[25] (3−7 equiv TBDMSCl, cat. DMAP, 2.5−3 equiv Et_3N, DMF, 25 °C, 16 h, 100%) followed by selective deprotection of the primary alcohol (HOAc−H_2O−THF, 3:1:1, 25 °C, 48 h) to provide **24**[25] (58%) along with recovered **23** (17%) and the diol **16** (23%), Scheme 3. Both **16** and **23** could be recycled, providing effective conversions to **24** exceeding 90%. Cyclization of **24** to afford the key intermediate **19** was accomplished either in one-step upon Mitsunobu activation[26] of the primary alcohol (1.5 equiv of DEAD, 1.5 equiv of Ph_3P, THF, 72%) or in higher conversion in two steps through formation of the mesylate **25**[25] (94%) followed by base-promoted closure to **19** (2.5 equiv of NaH, 97%).

A number of alternatives to the conversion of diol **16** to **19** were also examined. Although selective acetylation of the primary alcohol of **16** could be effectively accomplished (1.1 equiv of Ac_2O, 1.1 equiv of Et_3N, CH_2Cl_2, 0 °C, 73%), the subsequent protection of the secondary alcohol provided recovered **26**[27] or led to competitive acyl migration and a mixture of reaction products (Scheme 3). Similarly, selective mesylation of the primary alcohol to provide **27**[27] followed by attempted protection of the remaining secondary alcohol as its TBDMS ether or acetate **29**[27] analogous to related studies[23] failed to provide a useful route to **19**.

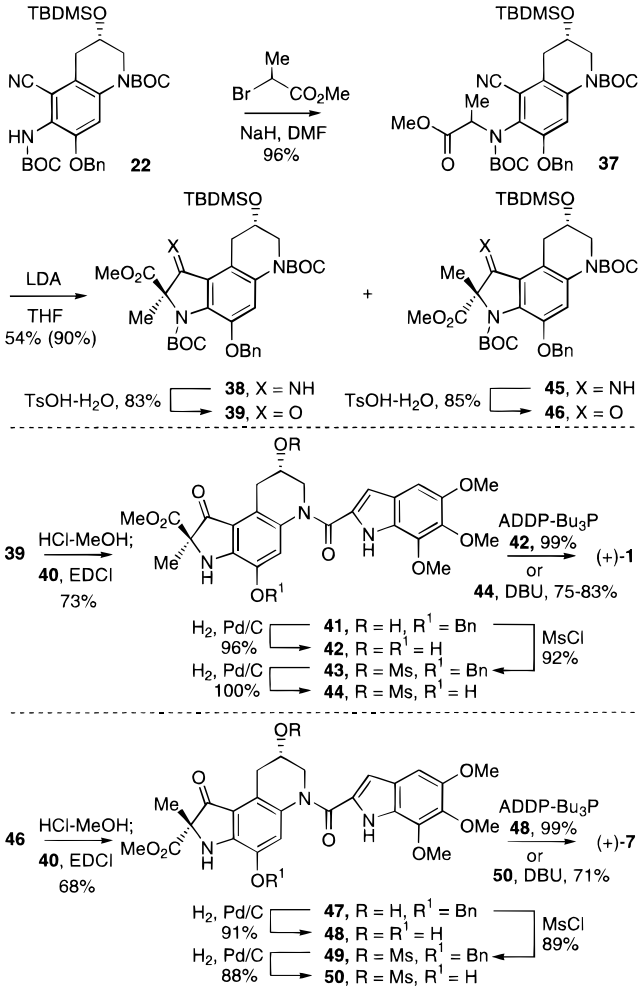
One potential significant improvement in the approach was investigated that utilizes the *p*-quinodiimine **32** incorporating the *N*-BOC protecting groups directly (Scheme 4). Conversion of **12** to **31**[25] via **30**[25] followed by oxidation to the *p*-quinodiimine **32**[25] provided the key substrate for this alternative approach. Regiospecific Lewis acid-catalyzed addition of allyltributyltin (0.5 of equiv of $BF_3{\cdot}OEt_2$, CH_2Cl_2, −78 °C, 2 h, 71%), catalytic dihydroxylation of **33**[25] (cat. OsO_4, 2.0 equiv of NMMO, acetone−H_2O, 25 °C, 95%), and selective tosylation of the primary alcohol **34**[25] (1.1 equiv of Bu_2SnO, toluene, reflux, 4 h; 1.2 equiv of TsCl, cat. Et_3N, 25 °C, 10 h, 88%) to provide **35**,[25] formation of the TBDMS ether **36**[25] (1.5 equiv of TBDMSOTf, 2 equiv of 2,6-lutidine, CH_2Cl_2, −30 °C, 2 h, 82%), and base-promoted closure[25] (2.0 equiv of NaH, 95%) provided **22**. Despite the improvement, the preparative resolution of **19** proved remarkably simple on a semipreparative ChiralCel OD HPLC column (α = 2.30) and much more effective than that observed with **22** (α = 1.14). For the preparation of optically active **1** and **7**, both the Sharpless asymmetric dihydroxylation of **15** versus **33** as well as the resolution of **19** or the enrichment of the optical purity of (*S*)- or (*R*)-**19** versus **22** proved more effective and was adopted.

Employing optically active **19** (>99.9% ee), the nondiastereoselective synthesis of (+)-duocarmycin A (**1**) and *epi*-(+)-

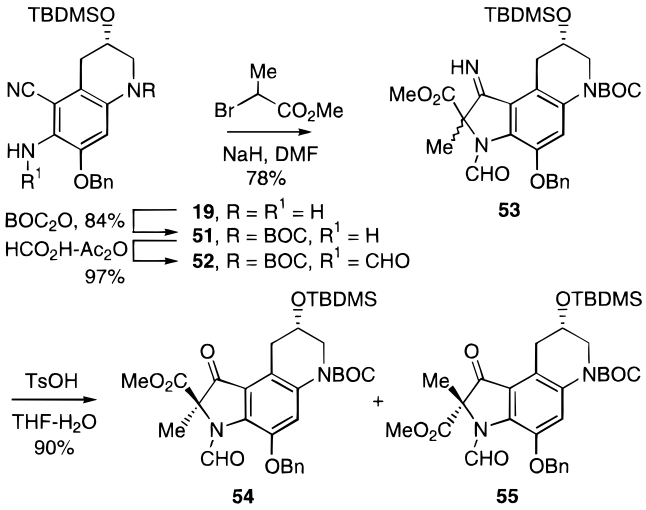
(26) Mitsunobu, O. *Synthesis* **1981**, 1.

(27) Diagnostic characterization is provided in the Supporting Information.

Scheme 5



Scheme 6



duocarmycin A (**7**) as well as their unnatural enantiomers was completed as shown in Scheme 5. *N*-Alkylation of **22** with methyl 2-bromopropionate (2 equiv of NaH, DMF, 0 °C, 0.5 h, 96%) provided **37**. Dieckmann-like condensation analogous to the efforts of Terashima[19] effected by treatment with LDA (3 equiv of, THF, −78 °C, 1 h, 54%, 90% based on recovered **37**) provided a separable 1:1 mixture of **38** and **45** (SiO_2, 20% EtOAc−hexane, $\alpha = 1.25$) epimeric at the C6 center. Interestingly, this readily reversible reaction was difficult to drive to completion and inevitably provided variable amounts of recovered starting material under the conditions examined. In the subsequent extension of this reaction to the *N*-formyl derivative or the Evans' acyl oxazolidinones, the ring closure product was more easily captured, presumably due to the decreased steric congestion of the product or the reduced acidity of the intermediate enolate, respectively. Mild hydrolysis of the imine (2−3 equiv of $TsOH{\cdot}H_2O$, $THF-H_2O$ 8:1, 0 °C, 2−3 h, 76−85%) conducted independently on the isomers or on the 1:1 mixture followed by chromatographic separation of **39** and **46** (SiO_2, 20% EtOAc−hexane, $\alpha = 1.33$) provided the key precursors to **1** and **7** without competitive *N*-BOC or TBDMS ether deprotection. Acid-catalyzed deprotection of **39** (4 M HCl-CH_3OH, 0 °C, 1 h), which served to remove both BOC and the TBDMS protecting groups, followed by coupling with 5,6,7-trimethoxyindole-2-carboxylic acid[10] (**40**, 73%) provided **41**. Removal of the benzyl ether (H_2, 10% Pd−C, 91−98%) followed by direct transannular spirocyclization of the alcohol **42** upon Mitsunobu activation (1.5 equiv of ADDP, 1.5 equiv of Bu_3P, C_6H_6, 50 °C 1 h, 99%) conducted under conditions analogous to those described by Natsume[28] provided (+)-duocarmycin A (**1**) identical in all respects with authentic material, $[\alpha]^{25}_D$ +291 (*c* 0.01, CH_3OH).[1] The intermediate alcohol **42**, duocarmycin D_1, has also been isolated from the culture broths of *Streptomyces* sp. DO-89, which produces (+)-duocarmycin A and constitutes a natural product in its own right.[7] Alternatively, treatment of **41** with MsCl (1.5 equiv, pyridine, 0 °C, 1 h, 89−92%) followed by hydrogenolysis of the benzyl ether **43** (H_2, 10% Pd-C, 88−100%) and spirocyclization effected by treatment of **44** with DBU[1] (83%) also provided (+)-duocarmycin A in conversions that proved more dependable on a small scale.

Similarly, acid-catalyzed deprotection of **46**[25] followed by coupling with 5,6,7-trimethoxyindole-2-carboxylic acid (**40**)[10] and the subsequent conversion of **47** to *epi*-(+)-duocarmycin A (**7**), $[\alpha]^{25}_D$ +155 (*c* 0.02, CH_3OH), was accomplished[25] as detailed for (+)-**1** (Scheme 5). By means of the same approach but employing (*R*)-**19** (>99.9% ee), the unnatural enantiomers of **1**, $[\alpha]^{25}_D$ −284 (*c* 0.025, CH_3OH), and **7**, $[\alpha]^{25}_D$ −156 (*c* 0.025, CH_3OH), were also prepared.

Similar efforts were conducted with the *N*-formyl derivative **52**,[25] which was obtained from **20** by BOC protection of the secondary amine (5 equiv of BOC_2O, CH_3CN, 90 °C, 16 h, 84%) followed by formylation of the primary amine of **51**[25] (HCO_2H-Ac_2O, 25 °C, 1 h, 93%). *N*-Alkylation of **52** with methyl 2-bromopropionate in the presence of excess NaH (3 equiv, DMF, 0 °C, 7 h, 78%) and subsequent *in situ* Dieckmann-like cyclization[19] provided **53**[25] directly (Scheme 6). Hydrolysis[25] of the imine **53** to the *β*-keto esters **54** and **55** was accomplished by exposure to TsOH (3 equiv, 4:1 $THF-H_2O$, 0 °C, 2 h, 90%). Although the intermediate **53** constituted a 1:1 mixture of diastereomers that was not separable, the two diastereomers **54** and **55** were separable (SiO_2, 30% EtOAc−hexane, $\alpha = 1.1$). Exhaustive acid-catalyzed deprotection of **54** or **55** was accomplished by treatment with saturated HCl−CH_3OH (25 °C, 5 h, 91%), and the resulting amine hydrochloride was immediately coupled with 5,6,7-trimethoxyindole-2-carboxylic acid (**40**)[10] employing EDCI (3 equiv, DMF, 25 °C, 48 h, 52%) or DEPC[29] (1.5 equiv, 1.5 equiv of Et_3N, THF, 70 °C, 16 h, 63%) to cleanly provide **41** and **47**. A number of alternative approaches were also examined, and a summary is provided in the Supporting Information.

Asymmetric Dihydroxylation of 15: Establishment of the Cyclopropane Absolute Stereochemistry. The overriding

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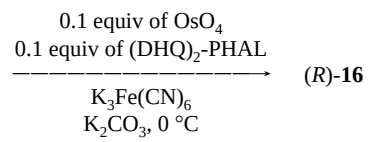
Table 1. Asymmetric Dihydroxylation of **15**

equiv of OsO_4	ligand (equiv)	condns[a] (solvent, time (h))	% yield	% ee
0.004	DHQD−CLB (0.25)	acetone−H_2O, 72	73	0
0.1	PHN−DHQ (0.1)	THF−H_2O, 48	60	0
0.1	MEQ−DHQ (0.1)	THF−H_2O, 48	12	0
0.04	$(DHQD)_2$−PYR (0.04)	THF−H_2O, 72	34	14 (*S*)
0.1	$(DHQD)_2$−PYR (0.1)	THF−H_2O, 48	69	36 (*S*)
0.1	$(DHQD)_2$−PYR (1.0)	THF−H_2O, 48	84	58 (*S*)
0.1	$(DHQ)_2$−PYR (0.5)	THF−H_2O, 48	73	39 (*R*)
0.1	$(DHQ)_2$−PYR$(OMe)_3$ (0.5)	THF−H_2O, 48	71	49 (*R*)
0.1	$(DHQ)_2$−PYR (0.1)	EtOAc−H_2O, 96	46	50 (*R*)
0.1	$(DHQD)_2$−AQN (0.2)	THF−H_2O, (4:1), 16	91	50 (*S*)
0.1	$(DHQD)_2$−AQN (0.2)	EtOAc−H_2O, 16	62	49 (*S*)
0.1	$(DHQ)_2$−PHAL (0.1)	THF−H_2O, 72	86	47 (*R*)
0.1	$(DHQ)_2$−PHAL (0.5)	THF−H_2O, 48	88	62 (*R*)
0.1	$(DHQ)_2$−PHAL (1.0)	THF−H_2O, 48	87	60 (*R*)
0.01[b]	$(DHQ)_2$−PHAL (0.1)	THF−H_2O, (4:1), 12	89	77 (*R*)
0.1	$(DHQ)_2$−PHAL (0.1)	EtOAc−H_2O, 48	58	74 (*R*)
0.1	$(DHQ)_2$−PHAL (0.5)	EtOAc−H_2O, 48	59	78 (*R*)
0.01[b]	$(DHQD)_2$−PHAL (0.1)	THF−H_2O, (4:1), 16	92	78 (*S*)

[a] 2:1 solvent−H_2O unless indicated otherwise. [b] Preparative scale.

Table 2. Solvent Effect on the Asymmetric Dihydroxylation[a]



solvent	% yield	% ee
t-BuOH−THF−H_2O	50	25
CH_3CN−H_2O	42	41
THF−H_2O	86	47
EtOAc−THF−H_2O	66	62
EtOAc−dioxane−H_2O	76	64
EtOAc−*t*-BuOMe−H_2O	46	66
EtOAc−toluene−H_2O	51	69
$ClCH_2CH_2Cl$−H_2O	59	69
EtOAc−acetone−H_2O	41	70
CH_2Cl_2−H_2O	68	73
EtOAc−H_2O	58	74
EtOAc−H_2O	59	78[b]
THF−H_2O (4:1)	89	77[c]

[a] 2:1 solvent−H_2O. [b] 0.5 equiv of ligand. [c] 4:1 THF−H_2O, 0.01 equiv of OsO_4, 0 °C, 16 h.

consideration in the adopted approach was its ability to provide either enantiomer of the key cyclopropane stereochemistry. Thus, complementary to the resolution of a prochiral diol such that all material could be employed to provide either or both enantiomers, our examination of methods of asymmetric synthesis was restricted to approaches that could provide access to both enantiomers. Since these elements are embodied in the Sharpless asymmetric dihydroxylation reaction,[23,30] it was especially attractive. Related efforts have been disclosed by Aristoff and conducted in the context of the synthesis of CBI-based analogs of CC-1065.[23] Representative results of the study of the asymmetric dihydroxylation of **15** are summarized in Tables 1 and 2 and proved far more interesting than anticipated or previously discussed.[23] The degree of asymmetric induction was assessed upon conversion of the diol **16** to the TBDMS ether **23** and analysis of the resulting enantiomers on an analytical ChiralCel OD HPLC column (0.46 × 25 cm, 13% *i*-PrOH−hexane, 0.3 mL/min flow rate, $t_R(S) = 31.5$ min, $t_R(R) = 27.5$ min, $\alpha = 1.15$). Without optimization of the conversions, an initial survey of the available ligands revealed that $(DHQ)_2$-PHAL/$(DHQD)_2$-PHAL[31] performed better than the $(DHQ)_2$-PYR/$(DHQD)_2$-PYR ligands,[32] which in turn were superior to their PHN, MEQ, or CLB[33] predecessors, Table 1.

In preliminary studies, the best results were obtained as the amount of ligand was increased, and because of the insolubility of **15** in *t*-BuOH, alternative solvent systems were also examined. The reaction was found to exhibit a large solvent dependency effecting the chemical conversion and a modest solvent dependency on the degree of asymmetric induction. Consequently, the effect of the solvent was examined in detail, and representative results are summarized in Table 2. In general, both the chemical yields and the ee's were best in the solvent systems in which **15** was most soluble (*i.e.*, 4:1 > 2:1 THF−H_2O), and the modest chemical conversions in many of the solvent systems may be attributed to the low solubility of **15** rather than slow osmate ester hydrolysis. Under the best conditions, dihydroxylation of **15** in the presence of $(DHQD)_2$-PHAL (0.1 equiv, 0.01 equiv of OsO_4, 3 equiv of $K_3Fe(CN)_6$, 3 equiv of K_2CO_3, 0.4 equiv of $CH_3SO_2NH_2$, 4:1 THF-H_2O, 16 h, 25 °C, 89−92%, 78% ee) provided (*S*)-**16**, and the complementary ligand $(DHQ)_2$-PHAL provided the corresponding enantiomer (89%, 77% ee).

Moreover, the absolute configuration of the product derived from the asymmetric dihydroxylation of **15** proved to be opposite that predicted from established models.[30] This was first recognized upon taking the major enantiomer derived from the $(DHQ)_2$-PHAL-catalyzed reaction through the synthesis that provided the unexpected unnatural enantiomer about the cyclopropane. Similarly, the major enantiomer derived from the complementary $(DHQD)_2$-PHAL-catalyzed reaction unexpectedly provided the natural enantiomer series, and these observations were repeated several times, ensuring that the samples were not inadvertently switched. Since the absolute configuration of natural **1** was established by X-ray analysis of a heavy atom derivative (**4**)[4] and firmly supported by its DNA alkylation properties, this suggested that it was the sense of asymmetric induction in the dihydroxylation reaction, and not the natural product absolute configuration, that was in question. Consequently, the (*S*)-Mosher ester of the primary alcohol of the major enantiomer of **16** derived from the $(DHQD)_2$-PHAL-catalyzed reaction was prepared and subjected to X-ray structure analysis (Scheme 7).[34] Comparison of the relative configuration unambiguously established that it possesses the *S* absolute configu-

(30) Kolb, H. C.; VanNieuwenhze, M. S.; Sharpless, K. B. *Chem. Rev.* **1994**, *94*, 2483.

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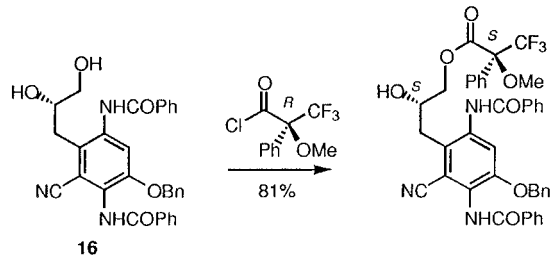
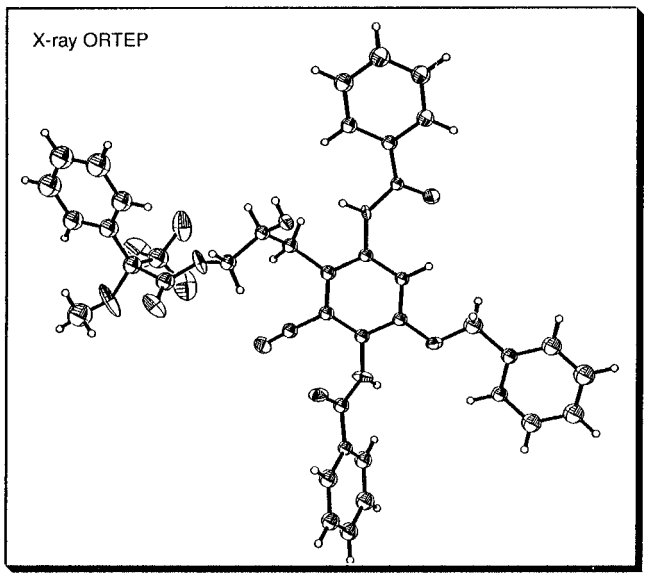
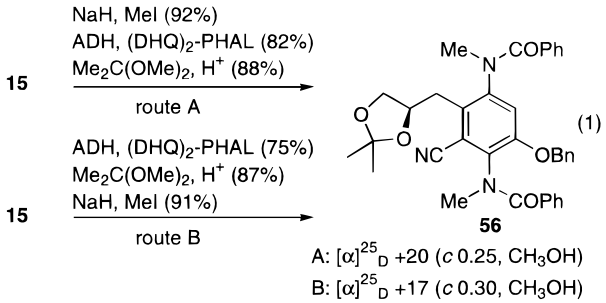
Scheme 7





ration contrary to expectations based on the established ADH models[35] but consistent with that required for synthesis of the natural product.

Similar observations were made with the substrate **33**, indicating that the degree and sense of asymmetric induction in the $(DHQ)_2$-PHAL-catalyzed reaction (0.02 equiv of OsO_4, 3 equiv of K_2CO_3, 3 equiv of $K_3Fe(CN)_6$, 2:1 *t*-BuOH−H_2O, 0 °C, 24 h, 93%, 56% ee, (*R*)-**34**) were not unique to the substrate **15** bearing the *N*-benzoyl protecting groups. In addition, the substrate containing a tertiary *N*-methylamide in place of the secondary amide of **15** exhibited the same sense and degree of asymmetric induction, indicating that the potential hydrogen bonding capabilities of the secondary amide **15** or carbamate **34** were not responsible for this reversal of enantioselectivity, eq 1.[25] Thus, the unusual reversal of the AD enantioselectivity



does not appear to be influenced by the obvious proximal olefin substituents of **15**, and such substrates may represent a more general class of olefins for which the AD enantioselectivity is reversed or more difficult to predict.

Resolution of Racemic 19 or Enrichment of the Enantiomeric Purity of Optically Active 19. The sensitivity of the biological assays to the contaminant enantiomers is exceptional, and even 1.0−0.1% of contaminant natural enantiomer in the unnatural enantiomier series complicates the conclusions that may be drawn from their examination. This was apparent in our initial studies conducted in collaboration with Terashima[13] on samples of the unnatural enantiomers of **1** and **7** derived from a diastereomeric derivatization and chromatographic resolution of racemic intermediates.[19] In these studies, the behavior of the unnatural enantiomers were masked by the dominant properties of the more potent and contaminating natural enantiomer (*ca.* 1%), and their characterization was limited to the establishment that they were ≥100× less effective than the corresponding natural enantiomer. Consequently, we examined protocols that could efficiently resolve our racemic intermediates or further enrich the enantiomeric purity of our optically active intermediates in a manner that could dependably provide materials that were >99.9% ee. This was effectively accomplished by direct chromatographic resolution[36−38] on a semipreparative ChiralCel OD HPLC column (2 × 25 cm). A series of intermediates were examined for potential resolution, and the results are summarized in Table 3. The resolution of the key intermediate **19** on a ChiralCel OD HPLC column proved remarkable, α = 2.30−1.94, and using even a semipreparative HPLC column (2 × 25 cm, 20−25% *i*-PrOH−hexane, 5 mL/min) 150 mg of racemic or 250 mg of optically enriched **19** (78% ee) could be resolved with a single injection and provided the enantiomers in >99.9% ee.

Table 3. Resolution on a ChiralCel OD Column[a]

agent	solvent	α
19	20% *i*-PrOH−hexane	2.30[b]
	25% *i*-PrOH−hexane	1.94[b]
22	3% *i*-PrOH−hexane	1.14[b]
23	13% *i*-PrOH−hexane	1.15

[a] 0.46 × 25 cm ChiralCel OD analytical HPLC column. [b] Semipreparative ChiralCel OD HPLCL column (2 × 25 cm).

Development of a Divergent Approach for the Diastereoselective Introduction of the Quaternary C6 Center. One key element of the approach to **1** or its epimer **7** rests with the introduction of the C6 quaternary center with control of its relative and absolute stereochemistry. Given our interest in both diastereomers, this suggested that its introduction on a late stage intermediate might prove most effective. In preliminary studies,[39] we had examined a number of approaches and found that a diastereoselective Dieckmann-like condensation employing Evans' optically active *N*-acyloxazolidinones was especially effective. Moreover, the Evans auxiliaries proved to be the only substrates in the series examined in which the diastereomers were readily separable by chromatography.

In initial studies, we found that slow addition of **57** to a solution of LDA (1.2 equiv) at −78 °C over a period of 30 min provided **58** and **59** in excellent yield and diastereoselection (7:1), Scheme 8. Moreover, the major and minor diastereomers

(34) The atomic coordinates have been deposited with the Cambridge X-ray Data Centre and may be obtained, on request, from the Director, Cambridge X-ray Data Centre, University Chemical Lab, Lensfield Road, Cambridge, CB2 1EW, UK.

(35) For additional exceptions, see: Krysan, D. J. *Tetrahedron Lett.* **1996**, *37*, 1375. Hale, K. J.; Manaviazar, S.; Peak, S. A. *Tetrahedron Lett.***1994**, *35*, 425. Vanhessche, K.; Sharpless, K. B. Unpublished results. With optically active substrates and double diastereoselection: Carreira, E. M.; DuBois, J. *J. Am. Chem. Soc.* **1994**, *116*, 10825. Iwashima, M.; Kinsho, T.; Smith, A. B., III. *Tetrahedron Lett.* **1995**, *36*, 2199. Krysan, D. J.; Rockway, T. W.; Haight, A. R. *Tetrahedron: Asymmetry* **1994**, *5*, 625.

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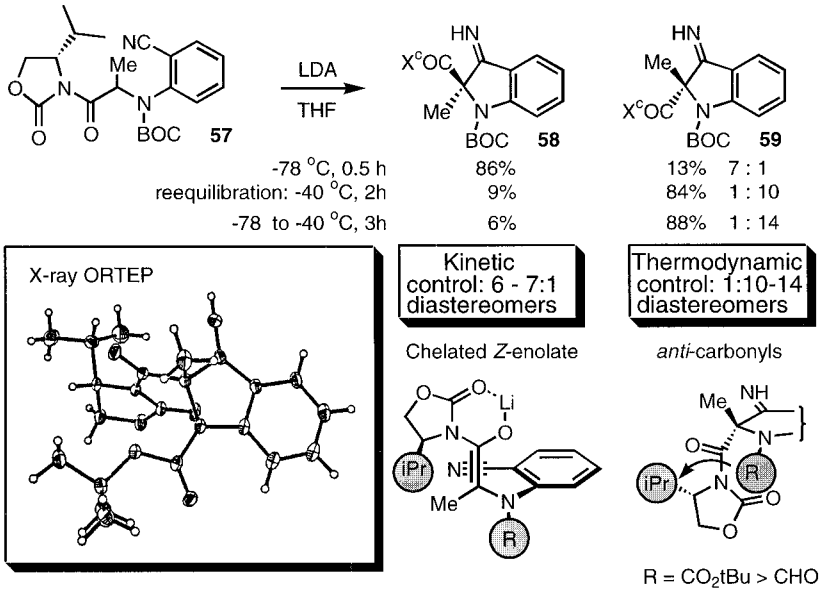
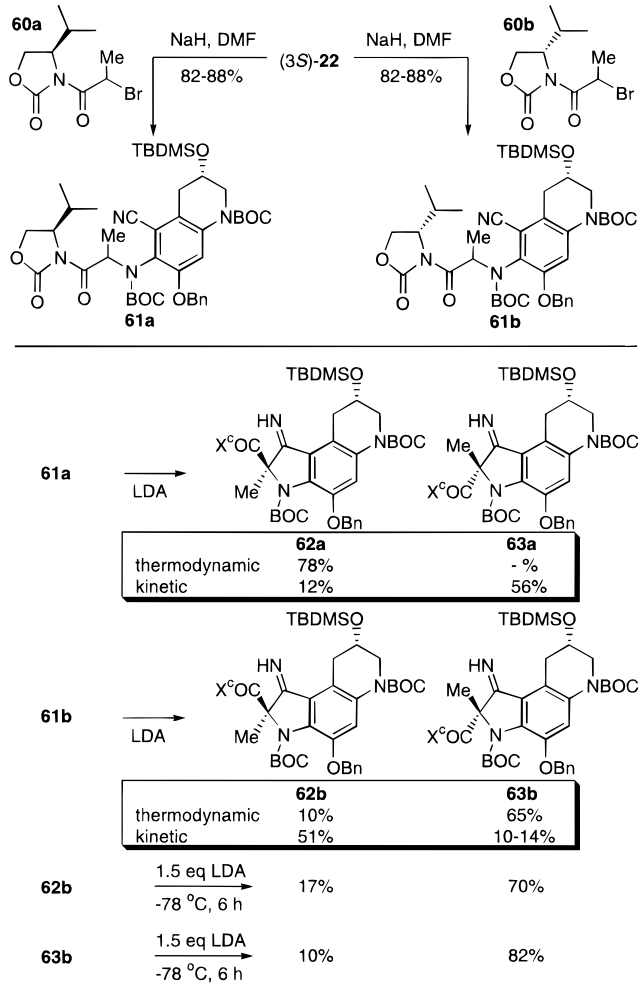
Scheme 8



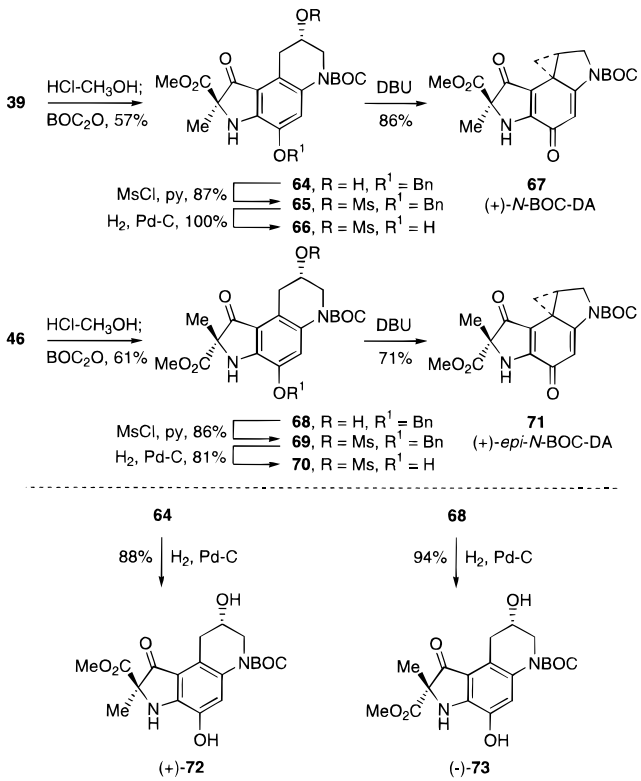
proved to be easily separable by chromatography (R_f = 0.40 and 0.78, α = 1.95; SiO_2, 50% EtOAc−hexane), and the desired diastereomer (2*R*)-**58** was isolated in 86% yield (>99.9% de). A single-crystal X-ray structure determination of the minor diastereomer (2*S*)-**59** established the relative and absolute stereochemistry. We also noted in this work that the diastereoselectivity of the reaction was dependent on the reaction conditions. Both reverse addition or addition over shorter time periods and at higher reaction temperatures led to diminished or even reversed diastereoselection. We now report that under kinetically controlled conditions, the reaction of **57** provides predominately **58** (7:1) as previously detailed, whereas under the thermodynamic conditions (1.5 equiv of LDA, THF, −40 °C, 3 h, 92−94%) of a reversible reaction it provides near-exclusive generation of **59** (14:1).[25] Establishment that **58** constitutes the kinetic product and **59** is the thermodynamic product of a reversible ring closure was obtained by resubjecting **58** to the reaction conditions (1.1 equiv of LDA, −40 °C, 2 h) with equilibration to **59** (10:1).[25] The stereochemical outcome is consistent with the derivation of the kinetic product from the reaction of a chelated (*Z*)-enolate, while the latter equilibration provides the most stable of the two possible diastereomers (ΔE = 0.76 and 0.41 kcal/mol, MM2 and AM1). The latter result is dependent on the size of the adjacent *N*-protecting group and its interaction with the chiral auxiliary substituent upon adoption of a nonchelated anti-carbonyl conformation with the acyl carbonyl eclipsed with the C-6 methyl group, and this is highlighted nicely in the X-ray structure of **59** (Scheme 8).

Divergent, Enantioselective Total Syntheses of (+)-Duocarmycin A and (+)-*epi*-Duocarmycin A and Their Unnatural Enantiomers. With the technology in hand to control the absolute configuration of the C6 stereocenter, we directed our efforts to the completion of an asymmetric total synthesis of the natural product. The complemenary stereochemistry of the kinetic versus thermodynamic Dieckmann-like condensation provided the option of utilizing a single intermediate bearing the same Evans oxazolidinone for the preparation of both natural (+)-duocarmycin A and (+)-*epi*-duocarmycin A in addition to the anticipated use of intermediates bearing the enantiomeric oxazolidinone auxiliaries. Alkylation of (3*S*)-**22** with the oxazolidone **60a**[39] (1.5 equiv of NaH, DMF, 0 °C, 1 h, 88%) followed by Dieckmann-like condensation of **61a** under thermodynamically-controlled reaction conditions best achieved by addition of base to a cooled solution of the substrate (6 equiv of LDA, THF, −78 to −50 °C, 0.5 h) cleanly provided the 2*R*,8*S* diastereomer **62a** (78%) as the near-exclusive closure product (≥8−10:1) and a small amount of recovered **61a** (7%), Scheme 9. Alternatively, condensation of **61b** incorporating the enantiomer of the acyl oxazolidinone under kinetically controlled conditions achieved by slow addition of substrate to base rigorously maintained at −78 °C (4−6 equiv of LDA, THF, −78 °C, 10 min) provided predominately **62b** (51%) also possessing the desired 2*R*,8*S* stereochemistry. That the latter constitutes the kinetic product and the former the thermodynamic

Scheme 9



Scheme 10



product of a reversible reaction was established by resubjecting pure **62b** and **63b** to LDA treatment with equilibration to the thermodynamic product **63b**. Methanolysis of **62a** or **62b** (30 equiv of $LiOCH_3$, THF−CH_3OH, 0 °C, 0.5 h, 78−84%) provided (2*R*,8*S*)-**38**, and its conversion to (+)-duocarmycin A (**1**) was accomplished as detailed in Scheme 5.

Similarly, closure of **61b** under conditions of thermodynamic control or condensation of **61a** under kinetic control where product equilibration is minimized provided **63b** (65%) or **63a** (56%), respectively, possessing the 3*S*,6*S* stereochemistry. Their methanolysis and conversion to *epi*-(+)-duocarmycin A (**7**) was accomplished as detailed in Scheme 5. Notably, the diastereomeric pairs **62**−**63a** (SiO_2, $\alpha = 1.40$, 20% EtOAc−hexane) and **62**−**63b** (SiO_2, $\alpha = 2.30$, 30% EtOAc−hexane) were readily separable by chromatography, thereby providing the pure diastereomers (>99.9% de). Thus, the diastereoselective preparation of **1** or its epimer **7** was achieved based on a divergent Dieckmann-like condensation in which either C6 absolute configuration could be introduced through choice of kinetic or thermodynamic reaction conditions on the same intermediate *or* through choice of complementary auxiliaries. By means of the same approach but employing (3*R*)-**22**, the unnatural enantiomers of **1** and **7** were also prepared.

Preparation of (+)-*N*-BOC-DA, (+)-*epi*-*N*-BOC-DA and Their Unnatural Enantiomers. In order to permit the examination of key partial structures of **1** and their unnatural enantiomers as well as to provide samples key to the examination of their functional reactivity and reaction regioselectivity, (+)-*N*-BOC-DA (**67**) and 6-*epi*-(+)-*N*-BOC-DA (**71**)[25] were prepared as detailed in Scheme 10. Exhaustive deprotection of **39** or **46**[25] (5 M HCl-CH_3OH, 0 °C, 1 h), which served to remove both *N*-BOC and TBDMS protecting groups, followed by immediate reprotection of the more reactive secondary amine (3 equiv of BOC_2O, THF, reflux, 2 h, 61−57%) provided **64** and **68**,[25] respectively. Analogous to the final steps in the preparation of **1** and **7**, activation of the secondary alcohol toward spirocyclization through formation of the mesylate, hydrogenolysis removal of the benzyl ether, and final transannular spirocyclization provided (+)-*N*-BOC-DA (**67**), $[\alpha]^{25}_D$ +183 (*c* 0.01, THF), and 6-*epi*-(+)-*N*-BOC-DA (**71**), $[\alpha]^{25}_D$ +73 (*c* 0.02, THF). In a similar fashion, the unnatural enantiomers of **67**, $[\alpha]^{25}_D$ −190 (*c* 0.02, THF), and **71**,[25] $[\alpha]^{25}_D$ −67 (*c* 0.01, THF), were also prepared.

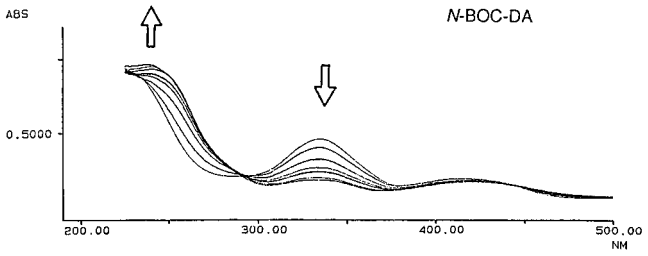


Figure 2. Solvolysis study (UV spectra) of *N*-BOC-DA in 50% CH_3OH−aqueous buffer (pH 3.0, 4:1:20 (v/v/v) 0.1 M citric acid, 0.2 M Na_2HPO_4, and H_2O, respectively). The spectra were recorded at regular intervals, and only a few are shown for clarity. Time intervals: 1, 0 h; 2, 3 h; 3, 6 h; 4, 9 h; 5, 12 h; 6, 24 h; 7, 36 h; 7, 48 h.

Finally, in conjunction with efforts to establish the stereochemistry of ring expansion solvolysis products obtained with **67**, both enantiomers of the diastereomeric alcohols **72** and **73** were prepared by hydrogenolysis of **64** and **68**.

Solvolysis Reactivity. Two characteristics of the alkylation subunits have proven important in the studies to date. The first is the stereoelectronically-controlled acid-catalyzed ring opening of the activated cyclopropane, which dictates preferential addition of a nucleophile to the least substituted cyclopropane carbon. The second is the relative rate of acid-catalyzed solvolysis, which has been found to accurately reflect the functional reactivity of the agents and to follow a fundamental, direct relationship between solvolysis stability and in vitro cytotoxic potency.[36−42]

The reactivity of **67** was assessed by following the solvolysis spectrophotometrically by UV at pH 3 (50% CH_3OH−buffer, buffer = 4:1:20 (v/v/v) 0.1 M citric acid, 0.2 M Na_2HPO_4, H_2O), measuring both the disappearance of the long-wavelength absorption band at 336 nm and the appearance of a short wavelength band attributable to the solvolysis products (Figure 2). As expected, *N*-BOC-DA (**67**, $t_{1/2} = 11$ h, $k = 1.75 \times 10^{-5}$ s^{-1}) proved to be more reactive toward chemical solvolysis at pH 3 than the alkylation subunit of CC-1065, *N*-BOC-CPI ($t_{1/2} = 37$ h), or duocarmycin SA, *N*-BOC-DSA ($t_{1/2} = 177$ h), Table 4. Thus, *N*-BOC-DA exhibits a stability at pH 3 that is not so distinct from that of *N*-BOC-CPI but that is substantially less than that of *N*-BOC-DSA.

Thus, the rate of acid-catalyzed solvolysis of *N*-BOC-DA was found to be substantially faster than that of *N*-BOC-DSA, presumably due to the enhanced gain in delocalization energy upon aromatization as well as the electronic deactivation by the conjugated C6 methoxycarbonyl substituent with duocarmycin SA. The magnitude of these effects is significant (16×) and revealing. This closely follows the trends established for the relative biological potency and DNA alkylation efficiency of the agents where the chemically more stable agents exhibit the more effective properties. In addition, the observation of a

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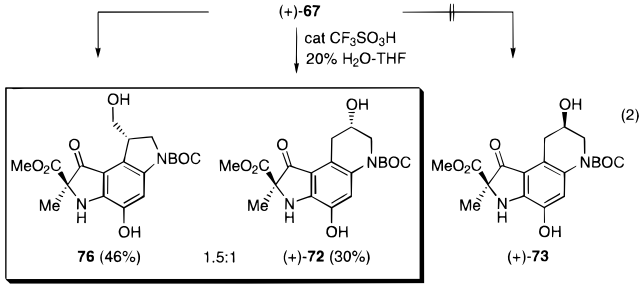
Table 4.

74, (+)-*N*-BOC-DSA	**75**, (+)-*N*-BOC-CPI	**67**, (+)-*N*-BOC-DA
$t_{1/2} = 177$ h, pH 3	$t_{1/2} = 37$ h, pH 3	$t_{1/2} = 11$ h, pH 3
regioselectivity: 6−4:1[43]	regioselectivity: 4:1	regioselectivity: 1.5:1
IC_{50} (L1210): 6 nM	IC_{50} (L1210): 300 nM	IC_{50} (L1210): 2 μM

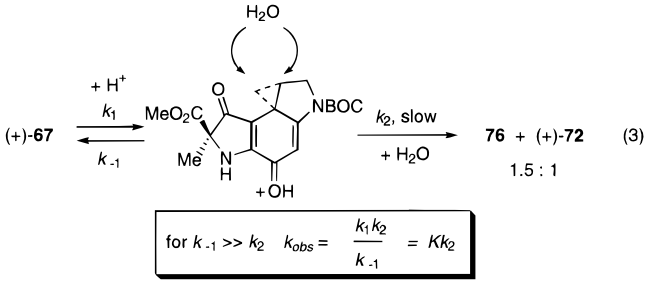
minor or trace guanine N3 alkylation with duocarmycin A that is not detected even under forcing conditions with duocarmycin SA may be attributed to this greater reactivity of duocarmycin A. Similarly, the greater ease of reversibility of the duocarmycin SA DNA alkylation reaction may be attributed to the lower degree of adduct stability resulting from its lower reactivity. Thus, consistent with this established relative reactivity, the duocarmycin A DNA alkylation reaction is less efficient, less selective among the available sites, and less reversible than that of duocarmycin SA.[14]

Solvolysis Regioselectivity and Mechanism of Acid-Catalyzed Nucleophilic Addition. As evidenced by the isolation and characterization of duocarmycins D_1 and D_2 and as observed in the studies of Saito,[44] the nucleophilic additions to the duocarmycin A system occur with both normal and abnormal ring expansion cleavage of the cyclopropane. An assessment of the acid-catalyzed additions to **67** was conducted and established that solvolysis preferentially occurs with cleavage of the C7b−C8 bond with addition of a nucleophile to the least substituted C8 cyclopropane carbon versus cleavage of the C7b−C8a bond with ring expansion and addition of a nucleophile to C8a. The latter cleavage would place a developing partial positive charge on a preferred secondary versus primary center and in preceding agents, this preference was overridden by the inherent stereoelectronic control of the reaction regioselectivity. However, this preference is greatly diminished with *N*-BOC-DA. Preparative solvolysis of **67** (20% H_2O−THF, 0.1 equiv of CF_3SO_3H, 25 °C, 24 h) cleanly provided both the normal solvolysis product **76** (46%) and the abnormal ring expansion solvolysis product (+)-**72** (30%) in a 1.5:1 ratio (eq 2). Moreover, no trace of the diastereomer (+)-**73** was detected,



indicating that under these conditions the ring expansion solvolysis occurs with clean inversion of the reacting center stereochemistry indicative of S_N2 addition to the activated cyclopropane. This is in complete agreement with the similarly unambiguous observations made in studies of the CBQ-based agents[42] and DSA-based agents[43] but contrasts the conclusions reached in a study of the CPI solvolysis where a free carbocation has been invoked to explain the observation of minor ring expansion products.[45] The results herein and those of the related studies[42,43] suggest that this later work should be reexamined using a more definitive experimental basis for assessing the stereochemical course of the reaction.

This proved consistent with kinetic studies of the acid-catalyzed nucleophilic addition conducted on related systems[37,38,45] where the rate of reaction exhibits a first-order dependence on both the acid concentration (pH) as well as the nucleophile, indicative of a mechanism involving rapid and reversible C4 carbonyl protonation followed by a slow, rate-determining S_N2 nucleophilic attack on the activated cyclopropane (eq 3).



To date, agents incorporating the CBI nucleus have exhibited the greatest regioselectivity (≥20:1)[37,38,40] for the cyclopropane ring-opening reaction including the exceptionally reactive F_2-CBI (≥9:1),[46] and more modest selectivity has been observed with CPI derivatives (*ca.* 4:1),[45] duocarmycin A (1.5−1:1),[7,44] duocarmycin SA (6−4:1),[43] or CBQ derivatives (3:2).[42] The distinguishing features controlling the regioselectivity of addition appear to be the stereoelectronic alignment of the two cyclopropane bonds available for cleavage and the relative reactivity of the agent. Within a class of agents whose cyclopropane alignment with the π-system would be expected to be very similar due to simple structural constraints, the solvolysis regioselectivity nicely follows the relative reactivity with the more stable agents providing the more selective reaction: *e.g.*, *N*-BOC-DSA (6−4:1)[43] > CPI (4:1)[45] > *N*-BOC-DA (3:2). However, this fails to hold true when comparing between classes of agents: *e.g.*, *N*-BOC-CBI (≥20:1) versus *N*-BOC-DSA (6−4:1). Thus, additional important factors contribute to this reaction regioselectivity. Like the comparisons made in the available structural studies of CPI,[47] DSA,[43] CBI,[40] MCBI,[37] CBQ,[42] and F_2CBI,[46] both the diminished regioselectivity of DSA relative to CBI and the very modest regioselectivity observed with the CBQ-based agents but maintained with the exceptionally reactive F_2CBI-based agents may be attributed to the relative extent of stereoelectronic alignment of the two possible cyclopropane bonds. In the cases where this structural information is available,[37,40,42,43,46,47] the degree of selectivity also reflects the relative degree of stereoelectronic alignment of the two available cyclopropane bonds, and this alone could account for the reaction regioselectivity.

DNA Alkylation Regioselectivity. The observation of exclusive adenine N3 addition to the C8 cyclopropane carbon in the DNA alkylation studies of duocarmycin A[9−13,46] is not consistent with expectations that the inherent acid-catalyzed

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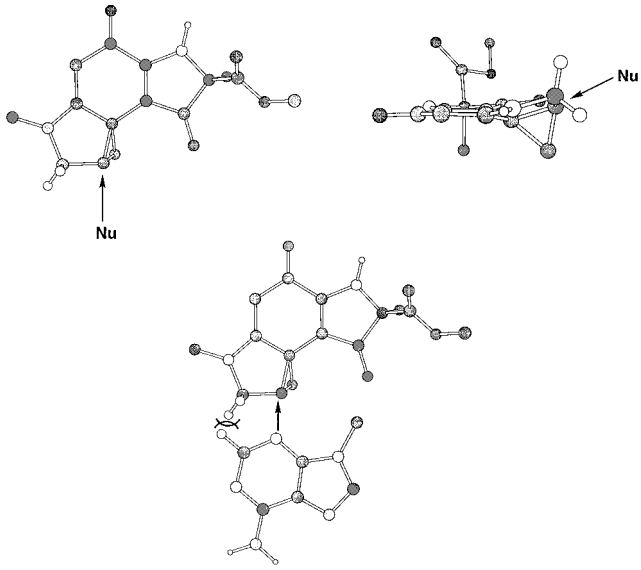

Figure 3. Top: top and side views of the nucleophile approach for abnormal ring expansion addition to the duocarmycin A activated cyclopropane illustrating the destabilizing torsional strain. Bottom: Abnormal adenine N3 addition confined to the reactant orientations taken from a model complex of duocarmycin A bound within the w794 DNA high affinity alkylation site illustrating the additional destabilizing steric interactions: C1−H_2/AdC2−H.

nucleophilic addition regioselectivity solely controls the DNA alkylation regioselectivity. This exclusive DNA alkylation regioselectivity was not only observed in our preceding studies with duocarmycin A (**1**), C_2, C_1,[9−12] or **7** and their enantiomers[13] but is general with all agents examined to date that undergo solvolysis with a mixed regioselectivity including duocarmycin SA (6:1),[14] the CPI-based agents (4:1 regioselectivity),[16,48,49] and the CBQ-based agents (3:2 regioselectivity).[46] Examination of each of these classes of agents has led only to detection of adducts derived from adenine N3 addition to the least substituted cyclopropane carbon. Moreover, each of these studies also quantitated the adduct formation and, in the case of duocarmycin A (86−92%),[12] duocarmycin SA (95−100%),[14] CC-1065 (> 85%),[48] and the CBQ-based agents (>75%),[42] established that the regioselectivity of the DNA alkylation reaction is greater than that of solvolysis. Although several explanations may be advanced for these observations,[42] the two most prominent are preferential adoption of binding orientations that favor normal adenine N3 addition (proximity effects) and the significant destabilizing torsional strain and steric interactions that accompany the abnormal addition especially when the reactants are restricted to the relative orientations found when bound in the minor groove. Figures illustrating these effects have been disclosed in our prior work,[42] and we would suggest that this latter subtle effect is most substantial (Figure 3). Consequently, the clean regioselectivity of the characteristic adenine N3 alkylation reaction benefits not only from stereoelectronic control but additional important subtle effects characteristic of a S_N2 reaction of a hindered nucleophile that complement the normally observed regioselectivity as well.

Cytotoxic Activity. With samples of the natural enantiomers of **1**, **7**, **67**, and **71** as well as their unnatural enantiomers in hand and of a high stereochemical purity (≥99.9% ee and de), their cytotoxic potency was established, Table 5. Consistent with the results of our prior studies with the Terashima samples,[13] (+)-duocarmycin was found to be 8× more potent than *epi*-(+)-duocarmycin A, and the unnatural enantiomers were 70−110× less potent than (+)-**1**. Similarly, (+)-*N*-BOC-DA was determined to be 4−5× more potent than *epi*-(+)-*N*-BOC-DA (**71**), and their unnatural enantiomers were ≥50× less potent than (+)-**67**.

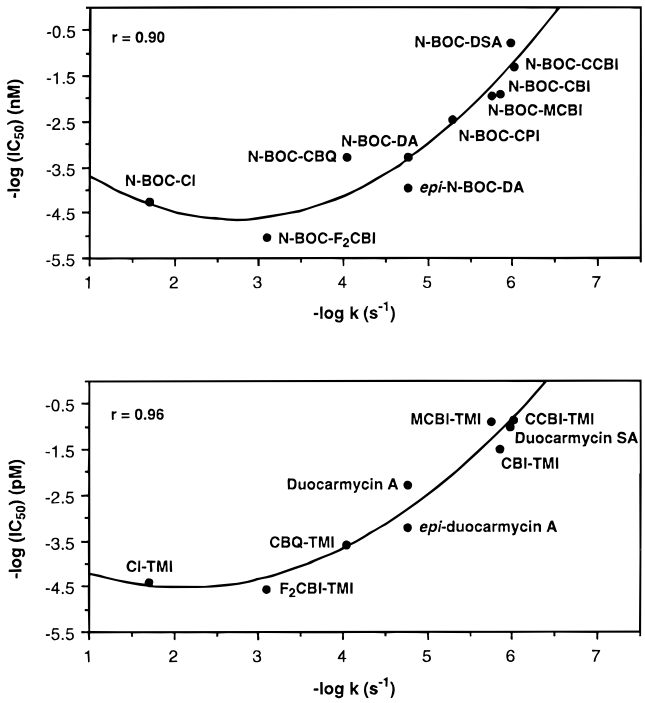

Figure 4.

Table 5. Cytotoxic Activity

agent	IC_{50} (L1210, nM)
(+)-**1**, (+)-duocarmycin A	0.2 nM
(+)-**7**, *epi*-(+)-duocarmycin A	1.6 nM
ent-(−)-**1**, *ent*-(−)-duocarmycin A	23 nM
ent-(−)-**7**, *ent*, *epi*-(−)-duocarmycin A	14 nM
(+)-**67**, (+)-*N*-BOC-DA	2 μM
(+)-**71**, *epi*-(+)-*N*-BOC-DA	9 μM
(−)-**67**, *ent*-(−)-*N*-BOC-DA	100 μM
(−)-**71**, *ent*,*epi*-(−)-*N*-BOC-DA	>100 μM

Despite the small differences in cytotoxic potency of the natural enantiomers but consistent with a well-established relationship,[5] the natural enantiomers follow the trends observed in prior studies that cover a large range of reactivities with the chemically more stable agents exhibiting the most potent activity (Figure 4).

Experimental Section

***N,N'*-Dibenzoyl-2-(benzyloxy)-6-cyano-1,4-benzoquinone Diimine (14).** $Pb(OAc)_4$ (15.49 g, 33.2 mmol) was added to a solution of **13**[25] (14.85 g, 33.2 mmol) at 0 °C, and the mixture was stirred for 4 h at 25 °C. The mixture was filtered through a Celite pad, and the filtrate was washed with saturated aqueous NaCl, dried ($MgSO_4$), and concentrated. Trituration of the residue with hexane gave **14** (12.4 g, 14.8 g theoretical, 84%) as yellow crystals: mp 118−120 °C (EtOAc, yellow plates); ^{1}H NMR (DMSO-d_6, 400 MHz) δ 8.00 (s, 1H), 7.89 (d, 1H, J = 0.8 Hz), 7.86 (d, 1H, J = 1.3 Hz), 7.70 (d, 1H, J = 7.5 Hz), 7.62−7.51 (m, 5H), 7.40 (t, 2H, J = 7.9 Hz), 7.22 (t, 1H, J = 7.4 Hz), 7.11 (t, 2H, J = 7.7 Hz), 6.80 (d, 2H, J = 7.4 Hz), 6.27 (br s, 1H), 4.90 (br s, 1H), 4.83 (br s, 1H); ^{13}C NMR (DMSO-d_6, 100 MHz) δ 178.5, 176.5, 155.9, 152.1, 146.3, 143.0, 134.1, 133.4, 133.3, 131.8, 131.2, 129.4, 129.0, 128.6, 128.5, 128.24, 128.15, 128.0, 120.3, 113.9,

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104.5, 71.1; IR (KBr) ν_{max} 3435, 3056, 2236, 1667, 1625, 1580, 1446, 1379, 1312, 1246, 1174, 1056, 1030, 841, 748, 728, 697 cm^{-1}. Anal. Calcd for $C_{28}H_{19}N_3O_3$: C, 75.49; H, 4.30; N, 9.43. Found: C, 75.39; H, 4.35; N, 9.47.

6-Allyl-3-(benzyloxy)-2,5-bis(benzoylamino)benzonitrile (15). A solution of **14** (5.50 g, 12.3 mmol) in anhydrous CH_2Cl_2 (55 mL) at −20 °C was treated with $BF_3{\cdot}Et_2O$ (0.76 mL, 6.18 mmol, 0.5 equiv). The mixture was stirred for 30 min and treated with allyltributyltin (4.21 mL, 13.6 mmol, 1.1 equiv). The reaction mixture was stirred for 3 h at −20 °C. Acetonitrile (250 mL) was added to the reaction mixture, the mixture was washed with hexane (2 × 100 mL), and the acetonitrile extract was concentrated *in vacuo* to afford crude **15**. The residue was crystallized from EtOAc to provide **15** (5.14 g, 6.02 g theoretical, 85%, typically 83−89%) as a white solid: mp 247.5−249.0 °C (CH_3OH, white needles); ^{1}H NMR (DMSO-d_6, 400 MHz) δ 10.28 (s, 1H), 10.17 (s, 1H), 7.90−8.00 (m, 4H), 7.42−7.65 (m, 9H), 7.25−7.35 (m, 3H), 5.85 (m, 1H), 5.17 (s, 2H), 5.00 (dd, 1H, J = 10.1, 1.5 Hz), 4.95 (dd, 1H, J = 17.1, 1.5 Hz), 3.58 (d, 2H, J = 6.0 Hz); ^{13}C NMR (DMSO-d_6, 100 MHz) δ 166.0, 153.0, 137.0, 136.7, 136.4, 134.6, 133.9, 133.7, 132.0, 131.9, 131.1, 128.6, 128.5, 128.4, 128.2, 127.9, 127.7, 127.2, 127.0, 118.1, 116.4, 115.5, 114.2, 70.2, 33.9; IR (KBr) ν_{max} 3251, 2226, 1651, 1600, 1580, 1523, 1492, 1451, 1415, 1344, 1308, 1272, 1241, 1144, 1103, 1026 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 620.0955 (M + Cs$^+$, $C_{31}H_{25}N_3O_3$ requires 620.0950). Anal. Calcd for $C_{31}H_{25}N_3O_3$: C, 76.37; H, 5.17; N, 8.62. Found: C, 76.31; H, 5.24; N, 8.51.

3-(Benzyloxy)-2,5-bis(benzoylamino)-6-(2,3-dihydroxypropyl)-benzonitrile (16). A solution of **15** (5.14 g, 10.5 mmol) and *N*-methylmorpholine *N*-oxide (2.47 g, 21.1 mmol, 2 equiv) in 4:1 acetone-H_2O (300 mL) was treated with OsO_4 (0.393 M toluene solution, 5.36 mL, 2.11 mmol, 0.2 equiv) and stirred overnight at 25 °C under N_2 in the dark. Sodium sulfite (1.0 g) was added, and the mixture was stirred for an additional 1 h. The mixture was diluted with saturated aqueous NaCl (300 mL) and extracted with EtOAc (3 × 150 mL). The combined organic extract was dried ($MgSO_4$) and concentrated *in vacuo*. The residue was crystallized from CH_3OH to afford **9** (5.22 g, 5.50 g theoretical, 95%) as a white solid: mp 231−232 °C (CH_3OH, white needles); ^{1}H NMR (DMSO-d_6, 400 MHz) δ 10.69 (s, 1H), 10.25 (s, 1H), 7.95−8.05 (m, 5H), 7.52−7.67 (m, 6H), 7.43 (br d, 2H, J = 6.6 Hz), 7.23−7.34 (m, 3H), 5.99 (br s, 1H), 5.17 (s, 2H), 5.05 (br s, 1H), 3.78 (m, 1H), 3.41 (m, 2H), 3.06 (dd, 1H, J = 14.4, 4.7 Hz), 2.89 (dd, 1H, J = 14.4, 8.8 Hz); ^{13}C NMR (DMSO-d_6, 100 MHz) δ 166.1, 165.0, 152.8, 137.6, 136.5, 134.0, 133.8, 132.2, 132.0, 128.8, 128.6, 128.4, 127.94, 127.91, 127.7, 127.4, 127.2, 126.8, 115.7, 114.7, 114.5, 72.9, 70.2, 65.3, 34.0; IR (KBr) ν_{max} 3421, 2933, 2226, 1656, 1605, 1580, 1518, 1415, 1349, 1308, 1272, 1241, 1149, 1097, 1072, 1021 cm^{-1}; FABHRMS (NBA) *m/z* 522.2029 (M + H$^+$, $C_{31}H_{27}N_3O_5$ requires 522.2029). Anal. Calcd for $C_{31}H_{27}N_3O_5$: C, 71.39; H, 5.22; N, 8.06. Found: C, 71.35; H, 5.52; N, 7.89.

Asymmetric Dihydroxylation of 15. A solution of OsO_4 (0.4 M in toluene, 0.32 mL, 0.127 mmol, 0.01 equiv), $K_3Fe(CN)_6$ (12.6 g, 38.1 mmol), K_2CO_3 (7 g), $CH_3SO_2NH_2$ (0.5 g, 5.0 mmol, 0.4 equiv), and $(DHQD)_2$-PHAL (643 mg, 1.27 mmol, 0.1 equiv) was added to a solution of **15** (6.2 g, 12.7 mmol) in THF−H_2O (4:1, 400 mL) at 0 °C, and the reaction mixture was stirred overnight at 25 °C. The reaction mixture was poured into EtOAc, and the organic layer was washed with aqueous 1 N HCl (2 × 100 mL), saturated aqueous $NaHCO_3$, and saturated aqueous NaCl, dried ($MgSO_4$), and concentrated. Trituration of the residue with Et_2O gave 6.1 g (92%) of **16**: mp 228−230 °C. The enantiomeric excess of **16** was determined upon conversion to **23** and analysis on a ChiralCel OD HPLC column (Daicel, 0.46 × 25 cm, eluent = 13% 2-propanol−hexane, flow rate = 0.3 mL/min). The effluent was monitored at 254 nm, and the enantiomers (3*R*)-**23** and (3*S*)-**23** were eluted with retention times of 27.5 and 31.5 min, respectively.

(*S*)-Mosher Ester of 16. (*R*)-MTPA-Cl (72 μL, 0.38 mmol) was added to a mixture of (*S*)-**16** (200 mg, 0.38 mmol, >90% ee derived from the $(DHQD)_2$-PHAL ADH) and Et_3N (54 μL, 0.57 mmol) in CH_2Cl_2 at 0 °C. After being stirred overnight at 25 °C, the reaction mixture was washed with 5% aqueous HCl, saturated aqueous $NaHCO_3$, and saturated aqueous NaCl, dried ($MgSO_4$), and concentrated. Chromatography (SiO_2, 1 × 20 cm, 33% EtOAc−hexane) followed by crystallization from EtOAc−hexane gave 226 mg (81%, >95% ee) of product. Recrystallization from CH_2Cl_2−hexane gave enantiomericallly pure material (>99% ee). Colorless plates for X-ray analysis were obtained by slow recrystallization from CH_2Cl_2-hexane: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.12 (s, 1H), 8.14 (s, 1H), 7.95−7.88 (m, 5H), 7.58−7.53 (m, 2H), 7.52−7.42 (m, 6H), 7.40−7.28 (m, 8H), 5.15 (d, 1H, J = 11.4 Hz), 5.10 (d, 1H, J = 11.4 Hz), 4.69 (dd, 1H, J = 2.2 Hz, 11.1 Hz), 4.40−4.30 (m, 2H), 3.500 (s, 3H); 3.498 (s, 1H), 3.15−3.01 (m, 2H); IR (neat) ν_{max} 3286, 2359, 1748, 1659, 1581, 1494, 1422, 1271, 1184, 1104, 1025 cm^{-1}; FABHRMS (NBA-NaI) *m/z* 738.7301 (M + H$^+$, $C_{41}H_{34}F_3N_3O_7$ requires 737.7293). The X-ray structure determination established the relative and *S*,*S* absolute stereochemistry.[34]

3-(Benzyloxy)-2,5-bis(benzoylamino)-6-[2-hydroxy-3-[(*p*-toluenesulfonyl)oxy]propyl]benzonitrile (17). A solution of **16** (2.20 g, 4.22 mmol) and Bu_2SnO (1.14 g, 4.60 mmol, 1.1 equiv) in 150 mL of toluene−THF (10:1) was warmed at reflux with azeotropic removal of H_2O with a Dean−Stark trap. After 6 h, 50 mL of solvent was distilled from the reaction vessel, and the solution was cooled to 0 °C. Et_3N (0.02 g, 0.2 mmol, 0.05 equiv) and *p*-TsCl (0.95 g, 5.0 mmol, 1.2 equiv) were added, and the reaction mixture was warmed to 25 °C and stirred overnight. The reaction mixture was quenched with the addition of 50 mL of saturated aqueous NaCl, and the aqueous layer was extracted with EtOAc (3 × 60 mL). The combined organic layer was dried (Na_2SO_4) and concentrated under reduced pressure. Flash chromatography (SiO_2, 50% EtOAc−hexane) afforded **17** (2.54 g, 2.85 g theoretical, 89%; typically 89−94%) as white crystals: mp 165−167 °C (CH_3OH, white needles); ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.91 (s, 1H), 8.12 (s, 1H), 7.96 (d, 2H, J = 7.0 Hz), 7.88−7.86 (m, 3H), 7.79 (d, 2H, J = 8.4 Hz), 7.59−7.43 (m, 6H), 7.33−7.28 (m, 7H), 5.13 (d, 1H, J = 11.3 Hz), 5.08 (d, 1H, J = 11.3 Hz), 4.31−4.29 (m, 1H), 4.24 (dd, 1H, J = 10.6, 2.6 Hz), 4.10 (dd, 1H, J = 10.6, 7.9 Hz), 3.56 (s, 1H), 3.06−2.96 (m, 2H), 2.43 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 166.3, 165.8, 151.6, 145.2, 137.2, 135.5, 134.3, 134.1, 133.6, 132.5, 130.0, 129.1, 128.8, 128.7, 128.6, 128.2, 127.9, 127.6, 127.4, 125.6, 125.1, 115.5, 115.1, 113.0, 112.7, 73.0, 71.0, 70.9, 32.8, 25.4; IR (film) ν_{max} 3280, 2929, 1667, 1601, 1515, 1422, 1360, 1272, 1176, 1096 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 808.1105 (M + Cs$^+$, $C_{38}H_{33}N_3O_7S$ requires 808.1094).

3-(Benzyloxy)-2,5-bis(benzoylamino)-6-[2-[(*tert*-butyldimethylsilyl)oxy]-3-[(*p*-toluenesulfonyl)oxy]propyl]benzonitrile (18). A solution of **17** (2.25 g, 3.33 mmol) in 100 mL of CH_2Cl_2 at 0 °C was treated with 2,6-lutidine (0.71 g, 6.6 mmol, 2 equiv) and TBDMSOTf (1.32 g, 5.00 mmol, 1.5 equiv), and the solution was stirred at 0 °C for 3 h. The reaction mixture was quenched with the addition of saturated aqueous $NaHCO_3$ and extracted with EtOAc (3 × 40 mL). The combined organic layer was dried (Na_2SO_4) and concentrated under reduced pressure. Flash chromatography (SiO_2, 17% EtOAc−hexane) afforded **18** (1.92 g, 2.60 g theoretical, 73%; typically 67−75%) as white needles: mp 143−146 °C (EtOAc, white crystals); ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.18 (s, 1H), 8.20 (s, 1H), 8.05−8.02 (m, 2H), 7.96 (s, 1H), 7.89 (d, 2H, J = 7.9 Hz), 7.84 (s, 1H), 7.79 (d, 2H, J = 8.3 Hz), 7.59−7.48 (m, 4H), 7.43 (t, 2H, J = 7.4 Hz), 7.33−7.27 (m, 6H), 5.19 (1H, d, J = 11.6 Hz), 5.15 (d, 1H, J = 11.6 Hz), 4.31−4.28 (m, 1H), 4.25 (dd, 1H, J = 9.1, 4.0 Hz), 4.06 (dd, 1H, J = 9.1, 2.1 Hz), 3.31 (dd, 1H, J = 14.7, 11.8 Hz), 2.91 (dd, 1H, J = 14.7, 2.8 Hz), 2.42 (s, 3H), 0.60 (s, 9H), −0.11 (s, 3H), −0.41 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 165.9, 165.4, 151.1, 145.3, 136.9, 135.4, 132.5, 132.3, 132.2, 130.1, 130.0, 129.1, 128.7, 128.4, 128.0, 127.6, 127.5, 127.4, 127.3, 126.0, 125.0, 124.9, 115.4, 112.7, 112.2, 72.5, 71.3, 70.6, 33.2, 25.5, 21.6, 17.7, −5.4, −5.5; IR (film) ν_{max} 3045, 2928, 1665, 1600, 1520, 1424, 1363, 1266, 1176, 1096 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 922.1933 (M + Cs$^+$, $C_{44}H_{47}N_3O_7SSi$ requires 922.1958).

1-Benzoyl-6-(benzoylamino)-7-(benzyloxy)-3-[(*tert*-butyldimethylsilyl)oxy]-5-cyano-1,2,3,4-tetrahydroquinoline (19). From 18. A solution of **18** (4.65 g, 5.89 mmol) in THF (130 mL) at 0 °C was treated with NaH (60% dispersion in oil, 0.47 g, 11.8 mmol, 2.0 equiv) in several portions over 15 min. After 2 h, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (50 mL), and the aqueous layer was extracted with EtOAc (4 × 50 mL). The combined organic layer was dried (Na_2SO_4) and concentrated *in vacuo*. Chromatography (SiO_2, 2 × 10 cm, 30% EtOAc−hexane) provided **19** (3.37 g, 3.63 g theoretical, 93%; typically 92−97%) as a white solid: mp 145−147 °C (CH_3OH, white needles); ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.86 (br d, 2H, J = 7.2 Hz), 7.74 (s, 1H), 7.35−7.59 (m,

8H), 7.27−7.32 (m, 3H), 7.13−7.18 (m, 2H), 6.71 (br s, 1H), 4.53 (s, 2H), 4.40 (m, 1H), 4.28 (br d, 1H, J = 12.8 Hz), 3.54 (dd, 1H, J = 12.8, 2.0 Hz), 3.24 (dd, 1H, J = 17.6, 5.1 Hz), 3.05 (dd, 1H, J = 17.6, 3.4 Hz), 0.84 (s, 9H), 0.10 (s, 3H), 0.03 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 171.1, 165.7, 149.5, 137.6, 135.6, 135.4, 133.5, 132.3, 130.7, 128.7, 128.5, 128.4, 127.6, 127.1, 126.3, 123.2, 115.0, 114.5, 111.5, 71.4, 64.6, 50.5, 35.5, 25.7, 18.1, −4.9; IR (KBr) ν_{max} 3292, 2933, 2851, 2215, 1656, 1600, 1580, 1492, 1390, 1328, 1256, 1210, 1174, 1092 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 750.1764 (M + Cs$^+$, $C_{37}H_{39}N_3O_4Si$ requires 750.1764). Anal. Calcd for $C_{37}H_{39}N_3O_4Si$: C, 71.93; H, 6.36; N, 6.80. Found: C, 71.96; H, 6.31; N, 7.04.

From 25. A suspension of NaH (387 mg, 60%, 9.68 mmol, 2.5 equiv) in THF (20 mL) at 0 °C under N_2 was treated with a solution of **25**[25] (2.76 g, 3.87 mmol) in THF (20 mL). The reaction mixture was stirred for 4 h at 25 °C. The reaction mixture was poured into ice-cold 10% aqueous HCl (100 mL) and extracted with EtOAc (3 × 80 mL). The combined organic extract was washed with saturated aqueous NaCl, dried ($MgSO_4$), and concentrated *in vacuo*. Chromatography (SiO_2, 15−30% EtOAc−hexane gradient elution) afforded **19** (2.32 g, 2.39 g theoretical, 97%) as a white solid identical in all respects to that described above.

Resolution of 19. A solution of **19** (150 mg of racemic or 250 mg of 89:11 3*S*:3*R*-**19** in 1 mL of CH_3OH) was separated on a ChiralCel OD (Daicel) semipreparative column (2 cm × 25 cm) using 20−25% 2-propanol−hexane eluent at a flow rate of 5 mL/min. The effluent was monitored at 254 nm, and the enantiomers (3*R*)-**19** and (3*S*)-**19** eluted with retention times of 32−48 and 74−93 min, respectively. HPLC analysis of the separated enantiomers (>95% recovery) indicated that both were >99.9% ee.

For (3*S*)-**19**: $[\alpha]^{20}_D$ −116.0 (*c* 0.50, CH_3OH).

For (3*R*)-**19**:$[\alpha]^{20}_D$ +116 (*c* 0.5, CH_3OH).

Anal. Calcd for $C_{37}H_{39}N_3O_4Si$: C, 71.93; H, 6.36; N, 6.80. Found: C, 71.76; H, 6.70; N, 6.55.

7-(Benzyloxy)-3-[(*tert*-butyldimethylsilyl)oxy]-1-(*tert*-butyloxycarbonyl)-6-[bis(*tert*-butyloxycarbonyl)amino]-5-cyano-1,2,3,4-tetrahydroquinoline (21). A solution of **19** (4.44 g, 7.21 mmol) in EtOH (28 mL) was treated with 280 mL of 98% NH_2NH_2 under Ar and the reaction mixture warmed at 140 °C in a sealed vessel for 20 h. The mixture was cooled, poured onto H_2O (600 mL), and extracted with EtOAc (3 × 200 mL). The organic phase was washed with saturated aqueous NaCl (150 mL), dried ($MgSO_4$), and concentrated *in vacuo*. Chromatography (SiO_2, 10−20% EtOAc−hexane) afforded **20** (1.92 g, 2.95 g theoretical, 65%), which was used immediately in the following reaction, and variable amounts of the corresponding free alcohol (45−30%). A solution of **20** (1.85 g, 4.52 mmol), BOC_2O (5.92 g, 27.1 mmol, 6 equiv), and DMAP (0.05 g, 0.45 mmol, 0.1 equiv) in THF (100 mL) was warmed at reflux for 2 h. The reaction mixture was cooled to 25 °C and concentrated *in vacuo*. PCTLC (SiO_2, 4 mm plate, 0−20% EtOAc−hexane gradient elution) provided **21** (3.05 g, 3.21 g theoretical, 95%): white amorphous solid; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.71 (s, 1H), 7.26−7.36 (m, 5H), 5.08 (s, 2H), 4.14−4.11 (m, 1H), 3.79 (dd, 1H, J = 2.8, 12.9 Hz), 3.49 (dd, 1H, J = 7.6, 12.9 Hz), 3.11 (dd, 1H, J = 5.6, 17.1 Hz), 2.80 (dd, 1H, J = 6.2, 17.1 Hz), 1.56 (s, 9H), 1.40 (s, 9H), 1.38 (s, 9H), 0.86 (s, 9H), 0.10 (s, 6H); IR (neat) ν_{max} 2978, 2931, 2229, 1801, 1763, 1708, 1599, 1489, 1369, 1253, 1154, 1102 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 842.2848 (M + Cs$^+$, $C_{38}H_{55}N_3O_8Si$ requires 842.2813).

(3*S*)-**21**: $[\alpha]^{25}_D$ −16.4 (*c* 0.15, CH_3OH).

(3*R*)-**21**: $[\alpha]^{25}_D$ +16.0 (*c* 0.33, CH_3OH).

7-(Benzyloxy)-3-[(*tert*-butyldimethylsilyl)oxy]-1-(*tert*-butyloxycarbonyl)-6-[(*tert*-butyloxycarbonyl)amino]-5-cyano-1,2,3,4-tetrahydroquinoline (22). From 21. A solution of **21** (1.05 g, 1.48 mmol) in CH_2Cl_2 (120 mL) at 0 °C was treated with TFA (0.84 g, 7.39 mmol, 5 equiv), and the solution was stirred for 2 h. The reaction was quenched by the slow addition of saturated aqueous $NaHCO_3$ (10 mL). The organic layer was separated, dried (Na_2SO_4), and concentrated *in vacuo*. PCTLC (SiO_2, 4 mm plate, 30% EtOAc−hexane) provided **22** (0.84 g, 0.93 g theoretical, 93%): white amorphous solid; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.66 (s, 1H), 7.25−7.45 (m, 5H), 6.34 (s, 1H), 5.08 (s, 2H), 4.15−4.16 (m, 1H), 3.74 (d, 1H, J = 12.7 Hz), 3.50 (dd, 1H, J = 7.6, 12.7 Hz), 3.15 (dd, 1H, J = 5.6, 17.2 Hz), 2.83 (dd, 1H, J = 6.2, 17.2 Hz), 1.54 (s, 9H), 1.52 (s, 9H), 0.89 (s, 9H), 0.12 (s, 6H); IR (KBr) ν_{max} 3272, 2930, 2256, 1691, 1598, 1501, 1461, 1392, 1368, 1331, 1253, 1149, 1104, 1064, 1004 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 742.2279 (M + Cs$^+$, $C_{33}H_{47}N_3O_6Si$ requires 742.2288). ChiralCel OD HPLC (2 × 25 cm, 5 mL/min, 3% *i*-PrOH−hexane): t_R = 42 min for (+)-(3*R*)-**22** and t_R = 48 min for (−)-(3*S*)-**22**.

(3*S*)-**22**: $[\alpha]^{25}_D$ −12.4 (*c* 0.5, CH_3OH).

(3*R*)-**22**: $[\alpha]^{25}_D$ +13.0 (*c* 0.1, CH_3OH).

From 36. A solution of **36**[25] (2.75 g, 3.52 mmol) in THF (100 mL) at 0 °C was treated with NaH (60% dispersion in oil, 0.28 g, 7.04 mmol, 2 equiv) in several portions over 15 min. After 1 h, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ and the aqueous layer was extracted with EtOAc (4 × 30 mL). The combined organic layer was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 4 mm plate, 20% EtOAc−hexane) provided **22** (2.04 g, 2.15 g theoretical, 95%) as an amorphous solid identical in all respects to that described above.

Methyl 2-[*N*-(*tert*-Butyloxycarbonyl)-*N*-[7-(benzyloxy)-3(*S*)-[(*tert*-butyldimethylsilyl)oxy]-1-(*tert*-butyloxycarbonyl)-5-cyano-1,2,3,4-tetrahydroquinolin-6-yl]amino]propionate ((−)-37). A solution of (−)-**22** (105 mg, 0.17 mmol) in DMF (5 mL) at 0 °C was treated with NaH (60% dispersion in oil, 14 mg, 2 equiv), and the solution was stirred for 0.5 h. 2-Bromopropionate (57 mg, 2 equiv) was added, and the solution was stirred for an additional 0.5 h at 0 °C. The reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (5 mL), and the aqueous layer was extracted with EtOAc (4 × 10 mL). The combined organic extract was washed with H_2O (2 × 10 mL), dried (Na_2SO_4), and concentrated under reduced pressure. PCTLC (SiO_2, 2 mm plate, 15% EtOAc−hexane) provided (−)-**37** (115 mg, 120 mg theoretical, 96%): ^{1}H NMR (DMSO-d_6, 400 MHz) δ 7.75−7.68 (m, 1H), 7.43−7.33 (m, 5H), 5.14−5.02 (m, 2H), 4.34−4.23 (m, 2H), 3.89−3.77 (m, 1H), 3.56−3.29 (m, 4H), 3.12−3.05 (m, 1H), 2.73−2.62 (m, 2H), 1.49−1.46 (m, 9H), 1.37−1.13 (m, 12H), 0.85−0.79 (m, 9H), 0.10−0.04 (m, 6H); IR (film) ν_{max} 2930, 1709, 1488, 1368, 1317, 1253, 1154, 1101 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 828.2625 (M + Cs$^+$, $C_{37}H_{53}N_3O_8Si$ requires 828.2656).

(−)-(3'*S*)-**37**: $[\alpha]^{25}_D$ −17 (*c* 0.3, CH_3OH).

ent-(+)-(3'*R*)-**37**: $[\alpha]^{25}_D$ +15 (*c* 0.2, CH_3OH).

Dieckmann Closure of 37. A solution of (−)-**37** (63 mg, 0.09 mmol) in THF (5 mL) at −78 °C was treated with LDA (1.35 mL of a 0.2 M solution in THF, 3 equiv). After 1 h at −78 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (1 mL), and the aqueous layer was extracted with EtOAc (4 × 2 mL). The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 2 mm plate, 15% EtOAc−hexane, α = 1.25) provided (+)-**38** (18 mg, 29% R_f = 0.4) and (−)-**45** (16 mg, 25% R_f = 0.5) as colorless oils and recovered starting material (25 mg, 40%).

(3*S*)-7-(Benzyloxy)-3-[(*tert*-butyldimethylsilyl)oxy]-6-[*N*-(*tert*-butyloxycarbonyl)-*N*-[1-[(4*R*)-2-oxo-4-isopropyloxazolidin-3-yl)carbonyl]ethyl]amino]-5-cyano-1,2,3,4-tetrahydroquinoline (61a). Prepared following the procedure detailed below: amorphous solid, 4:1 diastereomeric mixture: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.74 (s, 0.8 H), 7.67 (s, 0.2H), 7.46−7.25 (m, 5H), 5.80−5.70 (m, 1H), 5.09 (s, 1.6H), 5.08 (s, 0.4H), 4.48−4.08 (m, 3H), 3.88−3.70 (m, 1H), 3.50−3.38 (m, 1.8H), 3.16−3.10 (m, 1H), 2.84 (dd, 0.2H, J = 6.8 Hz, 16.3 Hz), 2.40−2.23 (m, 1H), 1.57 (s, 9H), 1.51 (s, 9H), 1.40−1.30 (m, 3H), 0.89 (s, 9H), 0.88 (br s, 6H), 0.12 (s, 3H), 0.11 (s, 3H); IR (neat) ν_{max} 3497, 3415, 2923, 2226, 1779, 1709, 1598, 1482, 1455, 1389, 1364, 1251, 1205, 1154, 748, 702 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3163 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184); $[\alpha]^{25}_D$ −12 (*c* 0.5, CH_3OH).

(3*S*)-7-(Benzyloxy)-3-[(*tert*-butyldimethylsilyl)oxy]-6-[*N*-(*tert*-butyloxycarbonyl)-*N*-[1-[((4*S*)-2-oxo-4-isopropyloxazolidin-3-yl)carbonyl]ethyl]amino]-5-cyano-1,2,3,4-tetrahydroquinoline (61b). A solution of (−)-(3*S*)-**22** (80 mg, 0.13 mmol) in DMF (5 mL) at 0 °C was treated with NaH (60% dispersion in oil, 10.4 mg, 0.26 mmol, 2 equiv). After 0.5 h at 0 °C, **60b**[39] (69 mg, 0.26 mmol, 2 equiv) was added, and the solution was stirred an additional 0.5 h. The reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (5 mL), and the aqueous layer was extracted with EtOAc (4 × 5 mL). The combined organic extract was washed with H_2O (2 × 5 mL), dried (Na_2SO_4), and concentrated *in vacuo*. PCTLC (SiO_2, 2 mm plate, 20% EtOAc-hexane) provided (−)-(3*S*,4'*S*)-**61b** (89 mg, 104 mg theoretical, 86%): amorphous solid, 3:2 diastereomeric mixture; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.07 (s, 0.6H), 7.67 (s, 0.4H), 7.45−7.25 (m, 5H), 6.30−

5.98 (m, 0.2H), 5.85−5.75 (m, 0.8H), 5.20−4.95 (m, 2H), 4.52−3.81 (m, 4H), 3.51−3.00 (m, 3H), 2.92−2.78 (m, 1H), 2.44−2.22 (m, 1H), 1.53 (s, 9H), 1.51 (s, 5.4H), 1.41 (s, 3.6H), 1.20−1.10 (m, 3H), 1.85−1.72 (m, 15H), 0.11 (s, 3H), 0.10 (s, 3H); IR (neat) ν_{max} 3497, 3416, 2922, 2222, 1782, 1709, 1599, 1490, 1456, 1389, 1364, 1251, 1205, 1155, 744, 701 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3152 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184).

(3*S*,4'*S*)-**61b**: $[\alpha]^{25}_D$ −17 (*c* 0.3, CH_3OH).

(3*R*,4'*R*)-**61b**: $[\alpha]^{25}_D$ +15 (*c* 0.2, CH_3OH).

Dieckmann Reaction of (−)-(3*S*,4'*R*)-61a. Kinetic conditions: a precooled (−78 °C) solution of (−)-**61a** (10 mg, 0.0126 mmol) in THF (0.2 mL) was added to a solution of LDA (0.5 M, 150 μL, 0.075 mmol) in THF (1 mL) at −78 °C over a period of 10 min. The reaction mixture was quenched with the addition of saturated aqueous NH_4Cl and extracted with EtOAc. The organic layer was washed with saturated aqueous NaCl, dried ($MgSO_4$), and concentrated. Purification of the residue by PTLC (SiO_2, 30% EtOAc−hexane) gave **62a** (1.2 mg, 12%) and **63a** (5.6 mg, 56%).

Thermodynamic conditions: LDA (0.5 M, 150 μL, 0.075 mmol) in THF was added to a solution of **61a** (10 mg, 0.0126 mmol) in THF (0.2 mL) at −78 °C. The mixture was stirred for 30 min below −50 °C before being quenched with the addition of saturated aqueous $NaHCO_3$ and extracted with EtOAc. The organic layer was washed with saturated aqueous NaCl, dried ($MgSO_4$), and concentrated. PTLC (SiO_2, 30% EtOAc−hexane) gave **62a** (7.8−8.1 mg, 78−81%).

(2*R*,8*S*)-4-(Benzyloxy)-3,6-bis(*tert*-butyloxycarbonyl)-8-[(*tert*-butyldimethylsilyl)oxy]-1-imino-2-methyl-2-[[(4*R*)-2-oxo-4-isopropyloxazolidin-3-yl]carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline ((−)-62a): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.07 (s, 1H), 7.55 (s, 1H), 7.41−7.25 (m, 5H), 5.00 (s, 2H), 4.45−4.20 (m, 4H), 4.12−4.01 (m, 1H), 3.23−3.18 (m, 1H), 3.01−2.96 (m, 1H), 2.90−2.86 (m, 1H), 2.56−2.55 (m, 1H), 1.66 (s, 3H), 1.52 (s, 9H), 1.31 (s, 9H), 0.92−0.88 (m, 6H), 0.88 (s, 9H), 0.08 (s, 3H), 0.07 (s, 3H); IR (neat) ν_{max} 2966, 2924, 1788, 1700, 1366, 1310, 1252, 1149 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3181 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184).

(2*S*,8*S*,4'*R*)-**62a**: $[\alpha]^{25}_D$ −35 (*c* 0.10, THF).

(2*S*,8*S*)-4-(Benzyloxy)-3,6-bis(*tert*-butyloxycarbonyl)-8-[(*tert*-butyldimethylsilyl)oxy]-1-imino-2-methyl-2-[[(4*R*)-2-oxo-4-isopropyloxazolidin-3-yl]carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline ((−)-63a): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.07 (s, 1H), 7.60 (s, 1H), 7.46−7.26 (m, 5H), 5.01 (s, 2H), 4.50−4.45 (m, 1H), 4.30−4.20 (m, 3H), 4.00−3.89 (m, 1H), 3.35−3.31 (m, 1H), 3.07−3.05 (m, 1H), 2.93−2.91 (m, 1H), 2.71−2.66 (m, 1H), 1.66 (s, 3H), 1.52 (s, 9H), 1.31 (s, 9H), 0.93−0.88 (m, 6H), 0.87 (s, 9H), 0.10 (s, 3H), 0.09 (s, 3H); IR (neat) ν_{max} 2966, 2924, 1785, 1699, 1365, 1309, 1251, 1148 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3179 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184).

(2*S*,8*S*,4'*R*)-**63a**: $[\alpha]^{25}_D$ −68 (*c* 0.05, THF).

Dieckmann Reaction of (−)-(3*S*,4'*S*)-61b. Kinetic Conditions. A solution of (−)-**61b** (42 mg, 0.053 mmol) in THF (0.5 mL) was added slowly via cannula to a solution of LDA (1 mL of a 0.2 M solution in THF, 4 equiv) at −78 °C. After 2 h, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc (3 × 2 mL). The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 1 mm plate, 25% EtOAc−hexane, α = 1.7) provided (+)-**62b** (21.4 mg, 51%, R_f = 0.35) and (+)-**63b** (5.9 mg, 14%, R_f = 0.60) as colorless oils.

Thermodynamic Conditions. A solution of (−)-**61b** (58 mg, 0.073 mmol) in THF (0.5 mL) at −78 °C was treated with LDA (1.1 mL of a 0.2 M solution in THF, 3 equiv). After 8 h at −78 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc (3 × 2 mL). The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 1 mm plate, 25% EtOAc−hexane) provided (+)-**62b** (5.8 mg, 10%) and (+)-**63b** (37.7 mg, 65%).

Reequilibration. A solution of (+)-**62b** (8 mg, 0.01 mmol) in THF (0.2 mL) at −78 °C was treated with LDA (0.10 mL of a 0.2 M solution in THF, 1.5 equiv). After 8 h at −78 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc (3 × 2 mL). The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 0.3 mm plate, 25% EtOAc−hexane) provided (+)-**62b** (1.3 mg, 17%) and (+)-**63b** (5.6 mg, 70%).

(2*R*,8*S*)-4-(Benzyloxy)-3,6-bis(*tert*-butyloxycarbonyl)-8-[(*tert*-butyldimethylsilyl)oxy]-1-imino-2-methyl-2-[[(4*S*)-2-oxo-4-isopropyloxazolidin-3-yl]carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline ((+)-62b): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.15 (br s, 1H), 7.50 (br s, 1H), 7.39 (d, 2H, *J* = 6.9 Hz), 7.33−7.25 (m, 3H), 5.04 (s, 2H), 4.49−4.46 (m, 1H), 4.25 (t, 1H, *J* = 8.8 Hz), 4.16 (dd, 1H, *J* = 9.0, 2.8 Hz), 4.14−4.10 (m, 1H), 3.88 (br d, 1H, *J* = 11.5 Hz), 3.44−3.40 (m, 1H), 3.33 (dd, 1H, *J* = 16.9, 6.0 Hz), 2.86 (dd, 1H, *J* = 16.9, 5.3 Hz), 2.55−2.48 (m, 1H), 1.70 (s, 3H), 1.51 (s, 9H), 1.27 (s, 9H), 0.91 (apparent t, 6H, *J* = 6.9 Hz), 0.86 (s, 9H), 0.11 (s, 3H), 0.10 (s, 3H); IR (film) ν_{max} 2956, 2917, 1784, 1697, 1362, 1311, 1249, 1151 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3162 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184).

(2*R*,8*S*,4'*S*)-**62b**: $[\alpha]^{26}_D$ +90 (*c* 0.08, THF).

ent-(2*S*,8*R*,4'*R*)-**62b**: $[\alpha]^{25}_D$ −87 (*c* 0.1, THF).

(2*S*,8*S*)-4-(Benzyloxy)-3,6-bis(*tert*-butyloxycarbonyl)-8-((*tert*-butyldimethylsilyl)oxy)-1-imino-2-methyl-2-[[(4*S*)-2-oxo-4-isopropyloxazolidin-3-yl]carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline ((+)-63b): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.08 (br s, 1H), 7.57 (br s, 1H), 7.44 (d, 2H, *J* = 7.0 Hz), 7.34−7.27 (m, 3H), 5.05 (s, 2H), 4.43−4.40 (m, 1H), 4.27−4.15 (m, 3H), 4.03−4.00 (m, 1H), 3.33−3.30 (m, 1H), 3.08−3.06 (m, 1H), 2.92 (dd, 1H, *J* = 16.8, 8.2 Hz), 2.57−2.53 (m, 1H), 1.69 (s, 3H), 1.51 (s, 9H), 1.30 (s, 9H), 0.90 (apparent t, 6H, *J* = 7.2 Hz), 0.89 (s, 9H), 0.11 (s, 3H), 0.10 (s, 3H); IR (film) ν_{max} 2964, 2923, 1785, 1698, 1364, 1308, 1251, 1149 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 925.3179 (M + Cs$^+$, $C_{42}H_{60}N_4O_9Si$ requires 925.3184).

(2*S*,8*S*,4'*S*)-**63b**: $[\alpha]^{26}_D$ +23 (*c* 0.02, THF).

ent-(2*R*,8*R*,4'*R*)-**63b**: $[\alpha]^{25}_D$ −20 (*c* 0.01, THF).

Methyl (2*R*,8*S*)-4-(Benzyloxy)-8-[(*tert*-butyldimethylsilyl)oxy]-3,6-bis(*tert*-butyloxycarbonyl)-1-imino-2-methyl-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline-2-carboxylate ((+)-38).[50] **From 62a.** A solution of **62a** (11 mg, 0.014 mmol) in THF−CH_3OH (4:1, 2 mL) at 0 °C was treated with $LiOCH_3$ (2 M in CH_3OH, 0.21 mL, 0.42 mmol, 3.0 equiv). After 1 h at 0 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (2 mL). The aqueous layer was extracted with EtOAc (4 × 2 mL), and the combined organic extract was dried (Na_2SO_4) and concentrated in vacuo. PCTLC (SiO_2, 0.3 mm plate, 30% EtOAc−hexane) provided (+)-**38** (7.9 mg, 9.6 mg theoretical, 82%): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.34 (br s, 1H), 7.52 (br s, 1H), 7.49 (d, 1H, *J* = 6.9 Hz), 7.38−7.26 (m, 3H), 5.12 (d, 1H, *J* = 11.2 Hz), 5.08 (d, 1H, *J* = 11.2 Hz), 4.15−4.09 (m, 1H), 3.98−3.95 (m, 1H), 3.68 (s, 3H), 3.40 (dd, 1H, *J* = 18.8, 6.2 Hz), 3.37−3.32 (m, 1H), 3.00−2.96 (m, 1H), 2.01 (br s, 1H), 1.72 (s, 3H), 1.51 (s, 9H), 1.35 (s, 9H), 0.87 (s, 9H), 0.10 (s, 3H), 0.09 (s, 3H); IR (film) ν_{max} 2928, 2845, 1743, 1697, 1501, 1367, 1249, 1146 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 828.2689 (M + Cs$^+$, $C_{37}H_{53}N_3O_8Si$ requires 828.2656).

(2*R*,8*S*)-**38**: $[\alpha]^{25}_D$ +75 (*c* 0.06, THF).

ent-(2*S*,8*R*)-**38**: $[\alpha]^{25}_D$ −76 (*c* 0.05, THF).

Methyl (2*R*,8*S*)-4-(Benzyloxy)-8-[(*tert*-butyldimethylsilyl)oxy]-3,6-bis(*tert*-butyloxycarbonyl)-2-methyl-1-oxo-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline-2-carboxylate ((+)-39).[50] A solution of (+)-**38** (10 mg, 0.014 mmol) in THF-H_2O (8:1, 0.5 mL) at 0 °C was treated with $TsOH\cdot H_2O$ (8.0 mg, 0.042 mmol, 3 equiv). After 2 h at 0 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc. The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PCTLC (SiO_2, 0.3 mm plate, 15% EtOAc−hexane) provided (+)-**39** (8.3 mg, 83%) as a white foam: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.68 (br s, 1H), 7.51 (d, 2H, *J* = 6.9 Hz), 7.36−7.25 (m, 3H), 5.14 (d, 1H, *J* = 12.4 Hz), 5.10 (d, 1H, *J* = 12.4 Hz), 4.13−4.07 (m, 1H), 3.91 (dd, 1H, *J* = 12.7, 2.8 Hz), 3.69 (s, 3H), 3.36 (dd, 1H, *J* = 12.7, 6.8 Hz), 3.34 (dd, 1H, *J* = 18.4, 5.4 Hz), 2.97 (dd, 1H, *J* = 18.4, 6.6 Hz), 1.71 (s, 3H), 1.52 (s, 9H), 1.40 (s, 9H), 0.86 (s, 9H), 0.10 (s, 3H), 0.09 (s, 3H); IR (film) ν_{max} 2928, 2860, 1762, 1746, 1707, 1503, 1363, 1333, 1253, 1146 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 829.2464 (M + Cs$^+$, $C_{37}H_{52}N_2O_9Si$ requires 829.2496).

(2*R*,8*S*)-**39**: $[\alpha]^{25}_D$ +103 (*c* 0.10, THF).

ent-(2*S*,8*R*)-**39**: $[\alpha]^{25}_D$ −107 (*c* 0.03, THF).

(50) Characterization for the diastereomeric series **45**−**50** and **68**−**70** is provided in the Supporting Information.

Methyl (2*R*,8*S*)-4-(Benzyloxy)-8-hydroxy-2-methyl-1-oxo-6-[(5,6,7-trimethoxyindol-2-yl)carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo-[3,2-*f*]quinoline-2-carboxylate ((+)-41).[50] **From 39.** (+)-**39** (4.0 mg, 5.7 μmol) was treated with 4 M HCl−CH_3OH (0.5 mL) at 0 °C for 1 h. The volatiles were removed under a stream of N_2, and the residue was dissolved in DMF (0.5 mL). EDCI (3.3 mg, 3 equiv) and **40**[10] (2.2 mg, 1.5 equiv) were added, and the solution was stirred for 4 h at 25 °C. PTLC (SiO_2, 10 × 20 cm × 0.25 mm, 80% EtOAc−hexane) provided (+)-**41** (2.6 mg, 73%): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.04 (s, 1H), 7.29−7.25 (m, 5H), 7.10 (s, 1H), 6.72 (s, 1H), 6.50 (s, 1H), 5.22 (s, 1H), 4.89 (s, 2H), 4.38−4.30 (m, 2H), 4.05 (s, 3H), 3.91 (s, 3H), 3.88−3.85 (m, 1H), 3.86 (s, 3H), 3.76 (s, 3H), 3.36 (dd, 1H, J = 19.2, 5.4 Hz), 3.19 (dd, 1H, J = 19.2, 2.9 Hz), 1.57 (s, 3H); IR (film) ν_{max} 3364, 2923, 1738, 1687, 1610, 1513, 1426, 1303, 1241, 1108 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 748.1275 (M + Cs^+, $C_{33}H_{33}N_3O_9$ requires 748.1271).

(2*R*,8*S*)-**41**: $[\alpha]^{25}_D$ +17 (*c* 0.10, THF).

ent-(2*S*,8*R*)-**41**: $[\alpha]^{25}_D$ −17 (*c* 0.05, THF).

From 54. A sample of **54**[35] (40.4 mg, 0.079 mmol) was treated with saturated HCl−CH_3OH (10 mL) at 25 °C for 5 h. The solvent was removed *in vacuo*, and the residue was diluted with EtOAc (30 mL), and the mixture was washed with saturated aqueous $NaHCO_3$ (20 mL), saturated aqueous NaCl (20 mL) and dried ($MgSO_4$), and concentrated *in vacuo*. Chromatography (SiO_2, 20−30% EtOAc−hexane gradient elution) afforded the free amine (27.4 mg, 30.1 mg theoretical, 91%) as a pale yellow solid. A solution of the free amine (27.4 mg, 0.072 mmol) and 5,6,7-trimethoxyindole-2-carboxylic acid (**40**, 19.8 mg, 0.079 mmol) in anhydrous THF (3 mL) was treated with diethyl cyanophosphonate (16.3 μL, 0.11 mmol, 1.5 equiv) and Et_3N (15.0 μL, 0.11 mmol, 1.5 equiv) at 0 °C under N_2. The reaction mixture was warmed at 70 °C for 16 h. The solvent was removed *in vacuo*. Chromatography (SiO_2, 50−80% EtOAc−hexane gradient elution) afforded **41** (27.8 mg, 44.1 mg theoretical, 63%) as a pale yellow solid identical in all respects to that described above.

This coupling reaction was also accomplished as follows. A solution of the free amine (3.9 mg, 0.01 mmol) and **40** (3.1 mg, 0.012 mmol, 1.2 equiv) in anhydrous DMF (0.5 mL) was treated with EDCI (5.9 mg, 0.03 mmol, 3 equiv) at 25 °C for 48 h under N_2. The solvent was removed *in vacuo*, and flash chromatography (SiO_2, 50−80% EtOAc−hexane gradient elution) afforded **41** (3.3 mg, 6.3 mg theoretical, 52%).

Methyl (2*R*,8*S*)-4,8-Dihydroxy-2-methyl-1-oxo-6-[(5,6,7-trimethoxyindol-2-yl)carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]-quinoline-2-carboxylate ((+)-42, Duocarmycin D_1).[50] A solution of (+)-**41** (11.8 mg, 19 μmol) in CH_3OH (1.0 mL) under H_2 was treated with 10% Pd−C (1.7 mg), and the suspension was stirred for 3 h at 25 °C. The reaction mixture was filtered through Celite and concentrated *in vacuo*. Chromatography (SiO_2, 30% EtOAc−hexane) provided (+)-**42** (9.7 mg, 10.1 mg theoretical, 96%; typically 92−98%) as a yellow solid: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.28 (s, 1H), 7.12 (s, 1H), 6.98 (s, 1H), 6.71 (s, 1H), 6.59 (s, 1H), 5.23 (s, 1H), 4.40−4.34 (m, 2H), 4.02 (s, 3H), 3.89 (s, 3H), 3.82 (s, 3H), 3.81−3.77 (m, 1H), 3.75 (s, 3H), 3.33 (dd, 1H, J = 19.0, 5.5 Hz), 3.19 (dd, 1H, J = 19.0, 2.8 Hz), 1.63 (s, 3H); IR (film) ν_{max} 3350, 2928, 1733, 1682, 1604, 1517, 1306, 1249, 1105 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 526.1846 (M + H^+, $C_{26}H_{27}N_3O_9$ requires 526.1826).

(2*R*,8*S*)-**42**: $[\alpha]^{25}_D$ +8 (*c* 0.04, THF).

ent-(2*S*,8*R*)-**42**: $[\alpha]^{25}_D$ −9 (*c* 0.04, THF).

Methyl (2*R*,8*S*)-4-(Benzyloxy)-8-[(methanesulfonyl)oxy]-2-methyl-1-oxo-6-[(5,6,7-trimethoxyindol-2-yl)carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline-2-carboxylate ((+)-43).[50] A solution of (+)-**41** (3.2 mg, 5.0 μmol) in pyridine (0.5 mL) was treated with MsCl (0.6 μL, 1.5 equiv), and the reaction mixture was stirred 1 h at 0 °C. The reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc. The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. Chromatography (SiO_2, 50−80% EtOAc−hexane gradient elution) provided (+)-**43** (3.3 mg, 92%): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.02 (s, 1H), 7.27−7.23 (m, 5H), 6.98 (s, 1H), 6.69 (s, 1H), 6.40 (s, 1H), 5.31−5.26 (m, 2H), 5.28 (s, 1H), 4.91 (d, 1H, J = 11.4 Hz), 4.87 (d, 1H, J = 11.4 Hz), 4.67−4.65 (m, 1H), 4.06 (s, 3H), 3.91 (s, 3H), 3.86 (s, 3H), 3.77 (s, 3H), 3.50−3.47 (m, 2H), 2.91 (s, 3H), 1.67 (s, 3H); IR (film) ν_{max} 3336, 292, 1740, 1692, 1618, 1512, 1459, 1310, 1236, 1171, 1104 cm^{-1}; FABHRMS (NBA-CsI) *m/z* (M + H^+, $C_{34}H_{35}N_3O_{11}S$ requires 694.2071).

(2*R*,8*S*)-**43**: $[\alpha]^{25}_D$ +11 (*c* 0.01, THF).

ent-(2*S*,8*R*)-**43**: $[\alpha]^{25}_D$ −11 (*c* 0.01, THF).

Methyl (2*R*,8*S*)-4-Hydroxy-8-[(methanesulfonyl)oxy]-2-methyl-1-oxo-6-[5,6,7-trimethoxyindol-2-yl)carbonyl]-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline-2-carboxylate ((−)-44).[50] A solution of (+)-**43** (3.3 mg, 5 μmol) in CH_3OH (0.5 mL) under H_2 was treated with 10% Pd−C (1 mg), and the suspension was stirred for 3 h at 25 °C. The reaction mixture was filtered through Celite and concentrated *in vacuo*. Chromatography (SiO_2, 30% EtOAc−hexane) provided (−)-**44** (2.9 mg, 100%) as a yellow solid: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.09 (s, 1H), 6.94 (s, 1H), 6.68 (s, 1H), 6.47 (s, 1H), 6.36 (br s, 1H), 5.29−5.23 (m, 2H), 4.71 (d, 1H, J = 11.7 Hz), 4.02 (s, 3H), 3.89 (s, 3H), 3.84−3.79 (m, 1H), 3.81 (s, 3H), 3.77 (s, 3H), 3.49−3.46 (m, 2H), 2.90 (s, 3H), 1.67 (s, 3H); IR (film) ν_{max} 3337, 2926, 2849, 1739, 1685, 1617, 1516, 1310, 1226, 1172, 1104 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 603.1535 (M^+, $C_{27}H_{29}N_3O_{11}S$ requires 603.1520).

(2*R*,8*S*)-**44**: $[\alpha]^{26}_D$ −7 (*c* 0.05, THF).

ent-(2*S*,8*R*)-**44**: $[\alpha]^{26}_D$ +6 (*c* 0.01, THF).

(+)-Duocarmycin A ((+)-1). From 44. A solution of (−)-**44** (8.5 mg, 14 μmol) in CH_3CN (1.0 mL) at 0 °C under Ar was treated with DBU (2 equiv, 4.2 μL, 28 μmol), and the solution was warmed to 25 °C and stirred for 1 h. The reaction mixture was diluted with EtOAc (10 mL), washed with cold 10% aqueous citric acid and saturated aqueous NaCl, dried ($MgSO_4$), and concentrated *in vacuo*. PTLC (SiO_2, 90% EtOAc−hexane, R_f = 0.65) provided **1** (5.3 mg, 7.1 mg theoretical, 75%; typically 75−83%): ^{1}H NMR (C_6D_6, 400 MHz) δ 9.22 (s, 1H), 7.65 (s, 1H), 6.68 (s, 1H), 6.43 (d, 1H, J = 2.3 Hz), 5.58 (s, 1H), 3.76 (s, 3H), 3.68 (s, 3H), 3.51 (s, 3H), 3.45 (d, 1H, J = 10.1 Hz), 3.22 (dd, 1H, J = 10.1, 5.2 Hz), 3.19 (s, 3H), 2.40−2.36 (m, 1H), 1.83 (dd, 1H, J = 7.6, 3.7 Hz), 1.41 (s, 3H), 0.42 (t, 1H, J = 4.6 Hz, partially obscured by H_2O); IR (film) ν_{max} 3321, 2935, 1743, 1683, 1622, 1386, 1303, 1274, 1235, 1199, 1110 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 508.1740 (M + H^+, $C_{26}H_{25}N_3O_8$ requires 508.1720).

Natural (+)-**1**: $[\alpha]^{26}_D$ +291 (*c* 0.01, CH_3OH).

ent-(−)-**1**: $[\alpha]^{26}_D$ −286 (*c* 0.025, CH_3OH).

From 42. A solution of **42** (9.7 mg, 0.018 mmol) in C_6H_6 (1 mL) was treated with Bu_3P (5.5 mg, 7 μL, 0.027 mmol, 1.5 equiv), and ADDP (7.0 mg, 0.027 mmol, 1.5 equiv) and the solution was warmed at 50 °C for 1 h under N_2. The volatiles were removed, and the residue was purified by chromatography (SiO_2, 1 × 4 cm, 80% EtOAc−hexane) to provide (+)-**1** (9.3 mg, 9.4 mg theoretical, 99%).

(+)-*epi*-Duocarmycin A ((+)-7). A solution of (−)-**50**[27] (1.40 mg, 2.32 μmol) in 0.5 mL of CH_3CN at 0 °C under Ar was treated with DBU (2 equiv, 23 μL of 0.20 M in CH_3CN) and the solution was warmed to 25 °C and stirred for 2 h. The reaction mixture was concentrated, and the crude product was purified by PTLC (5 × 20 cm × 0.25 mm, 90% EtOAc−hexane, R_f = 0.65) to provide **7** (0.84 mg, 71%): ^{1}H NMR (C_6D_6, 400 MHz) δ 9.16 (s, 1H), 6.95 (s, 1H), 6.66 (s, 1H), 6.41 (d, 1H, J = 2.1 Hz), 5.62 (s, 1H), 3.77 (s, 3H), 3.68 (s, 3H), 3.52 (s, 3H), 3.46 (d, 1H, J = 10.2 Hz), 3.34 (d, 1H, J = 10.2, 5.3 Hz), 3.20 (s, 3H), 2.41−2.39 (m, 1H), 1.86 (dd, 1H, J = 7.4, 3.8 Hz), 1.46 (s, 3H), 0.44 (t, 1H, J = 5.4 Hz, partially obscured by H_2O); IR (film) ν_{max} 3323, 2933, 1738, 1683, 1385, 1302, 1236, 1107 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 508.1744 (M + H^+, $C_{26}H_{25}N_3O_8$ requires 508.1720).

(+)-**7**: $[\alpha]^{26}_D$ +155 (*c* 0.015, CH_3OH).

ent-(−)-**7**: $[\alpha]^{26}_D$ −156 (*c* 0.025, CH_3OH).

Methyl (2*R*,8*S*)-4-(Benzyloxy)-6-(*tert*-butyloxycarbonyl)-8-hydroxy-2-methyl-1-oxo-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo[3,2-*f*]quinoline-2-carboxylate ((+)-64).[50] A sample of (+)-**39** (4.0 mg, 5.74 μmol) was treated with 5 M HCl-CH_3OH (0.5 mL), and the solution was stirred for 1 h at 0 °C. The volatiles were removed under a stream of N_2, and the residue was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL). The aqueous solution was extracted with EtOAc (3 × 1 mL), and the combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. The crude product[55] and BOC_2O (3.8 mg, 17 μmol, 3 equiv) were dissolved in THF (0.5 mL), and the solution was warmed at reflux for 2 h. The reaction mixture was concentrated, and the crude product was purified by PTLC (SiO_2, 10 × 20 cm × 0.25 mm, 50% EtOAc−hexane) to provide (+)-**64** (1.6 mg, 2.8 mg theoretical, 57%): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.45−7.33 (m, 6H),

5.28 (s, 1H), 5.09 (s, 3H), 4.27−4.24 (m, 1H), 3.89 (dd, 1H, J = 6.0, 13.0 Hz), 3.72 (s, 3H), 3.54 (dd, 1H, J = 2.0, 13.0 Hz), 3.27 (dd, 1H, J = 5.7, 18.8 Hz), 3.09 (dd, 1H, J = 4.2, 18.8 Hz), 1.72 (br s, 1H), 1.60 (s, 3H), 1.50 (s, 9H); IR (film) ν_{max} 3374, 2923, 2851, 1738, 1687, 1512, 1451, 1395, 1251, 1149 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 615.1134 (M + Cs$^+$, $C_{26}H_{30}N_2O_7$ requires 615.1107).

(2*R*,8*S*)-**64**: [α]$^{25}_D$ +106 (*c* 0.05, CH_3OH).

ent-(2*S*,8*R*)-**64**: [α]$^{25}_D$ −110 (*c* 0.05, CH_3OH).

Methyl (2*R*,8*S*)-4-(Benzyloxy)-6-(*tert*-butyloxycarbonyl)-8-[(methanesulfonyl)oxy]-2-methyl-1-oxo-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo-[3,2-*f*]quinoline-2-carboxylate ((+)-65).[50] A solution of (+)-**64** (6.0 mg, 12 μmol) in pyridine (0.1 mL) at 0 °C was treated with MsCl (1.9 μL, 25 μmol, 2 equiv). After 2 h at 0 °C, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ (0.5 mL), and the aqueous layer was extracted with EtOAc (3 × 1 mL). The combined organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PTLC (SiO_2, 20 × 20 cm × 0.25 mm, 50% EtOAc−hexane) provided (+)-**65** (5.7 mg, 6.7 mg theoretical, 85%; 85−87%) as a light yellow solid: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.45−7.34 (m, 6H), 5.16−5.13 (m, 1H), 5.14 (s, 1H), 5.11 (d, 1H, J = 11.2 Hz), 5.07 (d, 1H, J = 11.2 Hz), 4.31 (dd, 1H, J = 4.4, 12.9 Hz), 3.72 (s, 3H), 3.56 (d, 1H, J = 12.9 Hz), 3.38 (d, 2H, J = 6.8 Hz), 3.03 (s, 3H), 1.61 (s, 3H), 1.51 (s, 9H); IR (film) ν_{max} 2923, 1741, 1691, 1513, 1352, 1247, 1169 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 693.0851 (M + Cs$^+$, $C_{27}H_{32}N_2O_9S$ requires 693.0883).

(2*R*,8*S*)-**65**: [α]$^{25}_D$ +29 (*c* 0.07, CH_3OH).

ent-(2*S*,8*R*)-**65**: [α]$^{25}_D$ −28 (*c* 0.05, CH_3OH).

Methyl (2*R*,8*S*)-6-(*tert*-Butyloxycarbonyl)-4-hydroxy-8-[(methanesulfonyl)oxy]-2-methyl-1-oxo-2,3,6,7,8,9-hexahydro-1*H*-pyrrolo-[3,2-*f*]quinoline-2-carboxylate ((+)-66).[50] A solution of (+)-**65** (6.3 mg, 11.2 μmol) in CH_3OH (1.0 mL) under H_2 was treated with 10% Pd−C (1 mg), and the suspension was stirred for 2 h at 25 °C. The reaction mixture was filtered through Celite and concentrated *in vacuo*. PTLC (SiO_2, 20 × 20 cm × 0.25 mm, 75% EtOAc−hexane) provided (+)-**66** (5.3 mg, 5.3 mg theoretical, 100%): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.52 (s, 1H), 4.63 (s, 1H), 4.51−4.48 (m, 1H), 4.29 (dd, 1H, J = 4.0, 13.5 Hz), 3.55 (br s, 1H), 3.29 (d, 1H, J = 19.0 Hz), 3.18 (s, 3H), 2.97 (dd, 1H, J = 5.8, 19.0 Hz), 2.87 (d, 1H, J = 13.5 Hz), 1.99 (s, 3H), 1.54 (s, 9H), 1.45 (s, 3H); IR (film) ν_{max} 3350, 2926, 1739, 1690, 1513, 1359, 1248, 1169 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 603.0401 (M + Cs$^+$, $C_{20}H_{26}N_2O_9S$ requires 603.0413).

(2*R*,8*S*)-**66**: [α]$^{25}_D$ +52 (*c* 0.03, CH_3OH).

ent-(2*S*,8*R*)-**66**: [α]$^{25}_D$ −56 (*c* 0.03, CH_3OH).

(+)-*N*-BOC-DA ((+)-67). A solution of (+)-**66** (3.6 mg, 7.6 μmol) in CH_3CN (1.0 mL) cooled to 0 °C was treated with DBU (1.4 μL, 9.4 μmol), and the reaction mixture was warmed to 25 °C and stirred for 3 h. The mixture was diluted with EtOAc (10 mL), washed with 10% aqueous citric acid and saturated aqueous NaCl, and dried ($MgSO_4$). The solvent was removed *in vacuo*. PTLC (SiO_2, 20 × 20 cm × 0.25 mm, 80% EtOAc−hexane, or 33% acetone-$CHCl_3$) provided (+)-**67** (2.5 mg, 2.9 mg theoretical, 86%) as a pale yellow solid: ^{1}H NMR (C_6D_6, 400 MHz) δ 5.52 (s, 1H), 3.54 (br s, 1H), 3.22 (d, 1H, J = 11.3 Hz), 3.17 (s, 3H), 2.87 (dd, 1H, J = 4.7, 11.3 Hz), 2.29−2.25 (m, 1H), 1.71 (dd, 1H, J = 3.6, 7.6 Hz), 1.36 (s, 3H), 1.27 (s, 9H), 0.31 (t, 1H, J = 4.7 Hz); IR (film) ν_{max} 2923, 2851, 1725, 1677, 1559, 1446, 1385, 1251, 1148 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 507.0515 (M + Cs$^+$, $C_{19}H_{22}N_2O_6$ requires 507.0532).

Natural **67**: [α]$^{25}_D$ +183 (*c* 0.01, THF).

ent-**67**: [α]$^{25}_D$ −190 (*c* 0.02, THF).

(+)-6-*epi*-*N*-BOC-DA ((+)-71). Following the procedure described above, (−)-**70**[25] (0.85 mg, 1.8 μmol) provided (+)-**71** (0.51 mg, 0.68 mg theoretical, 71%): ^{1}H NMR (C_6D_6, 400 MHz) δ 5.59 (s, 1H), 3.51 (br s, 1H), 3.25 (d, 1H, J = 11.6 Hz), 3.16 (s, 3H), 2.98 (dd, 1H, J = 4.4, 11.6 Hz), 2.32−2.29 (m, 1H), 1.73 (dd, 1H, J = 3.7, 7.6 Hz), 1.43 (s, 3H), 1.25 (s, 9H), 0.31 (t, 1H, J = 4.3 Hz): IR (film) ν_{max} 2921, 2848, 1726, 1677, 1562, 1448, 1386, 1249, 1150 cm^{-1}; FABHRMS (NBA-CsI) *m/z* 507.0510 (M + Cs$^+$, $C_{19}H_{22}N_2O_6$ requires 507.0532).

(+)-71: [α]$^{25}_D$ +73 (*c* 0.02, THF).

ent-**71**: [α]$^{25}_D$ −67 (*c* 0.01, THF).

Aqueous Solvolysis Reactivity of 67. pH 3. *N*-BOC-DA (**67**, 100 μg) was dissolved in CH_3OH (1.5 mL) and mixed with pH 3 aqueous buffer (1.5 mL). The buffer contained 4:1:20 (v:v:v) 0.1 M citric acid, 0.2 M Na_2HPO_4, and H_2O, respectively. The solvolysis solution was sealed and kept at 25 °C protected from light. The UV spectrum was measured at regular intervals every 1 h during the first day and every 12 h for another week. The decrease in the long-wavelength absorption at 336 nm and the increase in the short-wavelength absorption were monitored. The solvolysis rate constant (k = 1.75 × 10^{-5} s^{-1}) and half-life ($t_{1/2}$ = 11 h) were calculated from data recorded at the long wavelength from the least-squares treatment (r = 0.98) of the slope of the plot of time versus $\ln[(A_f - A_i)/(A_f - A)]$.

Preparative Solvolysis of (+)-*N*-BOC-DA (67). A solution of (+)-**67** (0.35 mg, 0.93 μmol) in THF−H_2O (4:1, 0.2 mL) was cooled to 0 °C and treated with CF_3SO_3H (0.014 mg, 0.093 μmol, 0.1 equiv). The reaction mixture was warmed to 25 °C and stirred for 24 h. Saturated aqueous $NaHCO_3$ (0.5 mL) was added, and the aqueous layer was extracted with EtOAc (4 × 0.5 mL). The organic extract was dried (Na_2SO_4) and concentrated *in vacuo*. PTLC (SiO_2, 5 × 20 cm × 0.25 mm, 75% EtOAc) provided (+)-**72** (0.11 mg, 30%) identical in all respects with authentic material and **76** (0.17 mg, 46%). HPLC analysis (0.46 × 25 cm ChiralCel OD column, 10% *i*-PrOH−hexane, 1 mL/min) of the crude reaction mixture indicated the presence of **76** (t_R = 3.9 min, 60.5%), (+)-**72** (t_R = 7.8 min, 39.5%) and no (+)-**73** (t_R = 14.6 min, 0%) requiring that the ring expansion solvolysis reaction proceed by clean S_N2 addition of H_2O. HPLC analysis of authentic (+)-**72** and (+)-**73** under identicaconditions unambiguously established the t_R of the two possible ring expansion solvolysis products.

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Supporting Information Available: Diagnostic characterization of **26**, **27**, and **29**, a summary of alternative approaches explored, full experimental details and characterization for **9**−**13**, **23**−**25**, **30**−**36**, the diastereomeric series **45**−**50**, **51**−**55**, **56** (routes A and B), **58**, **59** and the diastereomeric series **68**−**70** and **72**−**73**, details of the X-ray structure determination of the (*S*)-Mosher ester of **16** (Scheme 7), and data employed in constructing Figure 4 (40 pages). See any current masthead page for ordering and Internet access instructions.

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