Total Synthesis of Bleomycin A_2 and Related Agents. 2. Synthesis of (–)-Pyrimidoblamic Acid, *epi*-(+)-Pyrimidoblamic Acid, (+)-Desacetamidopyrimidoblamic Acid, and (–)-Descarboxamidopyrimidoblamic Acid

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Abstract: Full details of concise syntheses of (–)-pyrimidoblamic acid (**1**), the authentic heterocyclic core of the bleomycin A_2 metal binding domain, as well as the key substructure analogs *epi*-(+)-pyrimidoblamic acid (**2**), (+)-desacetamidopyrimidoblamic acid (**3**), and (–)-descarboxamidopyrimidoblamic acid (**4**) are described. Key to the approach is the implementation of an inverse electron demand [4 + 2] cycloaddition reaction of 2,4,6-tris(ethoxycarbonyl)-1,3,5-triazine with 1-(dibenzylamino)-1-propyne or in situ generated 1,1-diaminopropene for the one-step preparation of an appropriately functionalized pyrimidine nucleus. The development and subsequent implementation of a diastereoselective imine addition reaction of optically active *N*-acyloxazolidinone enolates provided a stereocontrolled introduction of the pyrimidoblamic acid C2 acetamido side chain. Chemical studies which unambiguously establish and confirm the absolute configuration of the C2 acetamido side chain are detailed, and their extension to the synthesis of (–)-descarboxamidopyrimidoblamic acid (**4**) is described.

The bleomycins are a family of glycopeptide antitumor antibiotics possessing clinically useful activity thought to be mediated through their metal-dependent oxidative cleavage of duplex DNA[1] (Figure 1). Consequently, bleomycin A_2, its naturally occurring congeners, its semisynthetic derivatives and degradation products, and synthetic analogs have been the subject of extensive investigations in efforts to define the fundamental functional roles of its structural subunits. In the preceding article, we provided full details of concise syntheses of tri-, tetra-, and pentapeptide S as well as a series of structural analogs and the determination of their DNA binding properties.[2] Herein we provide full details of the synthesis of (–)-pyrimidoblamic acid (**1**),[3-5] *epi*-(+)-pyrimidoblamic acid (**2**),[4,5] (+)-desacetamidopyrimidoblamic acid (**3**),[4,6] and (–)-descarboxamidopyrimidoblamic acid (**4**) in efforts that represent a systematic exploration of the key structural features of the metal binding domain of bleomycin A_2.[1] Extensive studies from the Umezawa–Ohno laboratories have defined a number of seminal features of the pyrimidoblamic subunit through chemical derivatization or degradation of the natural product[7] and through chemical synthesis,[8] and a number of additional simplified metal binding domains have been evaluated in the laboratories of others.[4,5,9-13] Complementary to the efforts disclosed to date, our recent observation of the potentiating DNA cleavage effects of the C2

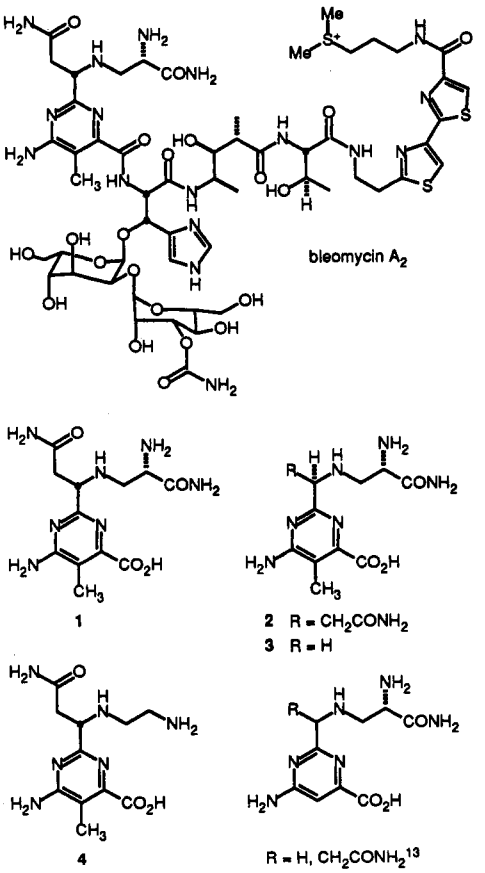

Figure 1.

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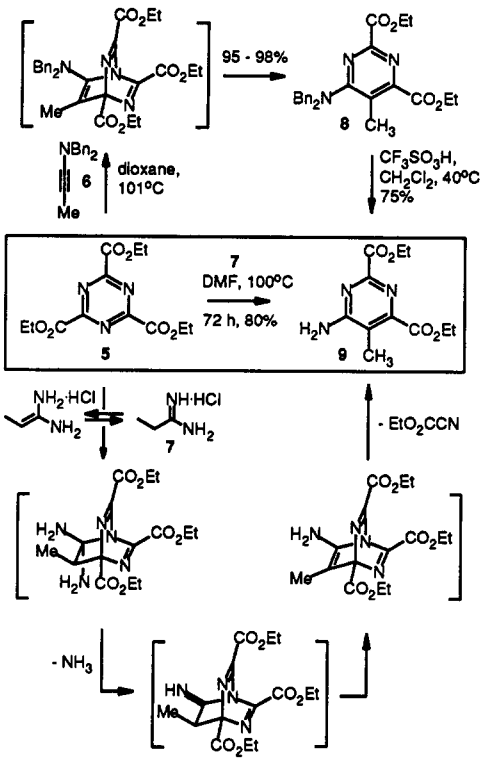
acetamido side chain of P-3A[13] and deglycobleomycin A_2[6] in addition to the proposed rapid metabolic inactivation of bleomycin A_2 through bleomycin hydrolase[7b,14] hydrolysis of the carboxamide have suggested fundamental modifications of the pyrimidoblamic acid subunit that have yet to be explored. Concurrent with such efforts, a study of the bleomycin A_2 analogs incorporating modified pyrimidoblamic acid subunits could be expected to permit an assessment of the role each substituent may play in contributing to metal chelation, O_2 activation, and DNA cleavage efficiency as well as the fundamental role the subunit may play in controlling the characteristic 5′-GC/GT DNA cleavage selectivity.[15] The diastereoselective synthesis of (−)-pyrimidoblamic acid (**1**) detailed herein concisely installs the two remaining acyclic stereogenic centers of the natural aglycone. Notably, this represents the first reported approach which permits control of the relative and absolute stereochemistry of the C2 acetamido side chain of **1** and does so in a concise manner readily adaptable to the concurrent preparation of analogs. In realization of efforts to prepare by chemical synthesis bleomycin A_2 analogs possessing deep-seated structural changes for subsequent evaluation, the extension of the studies to the synthesis of **2–4** is additionally detailed. Pertinent to the studies detailed herein, the C2 acetamido side chain and the carboxamido group of the bleomycins have been shown not to be intimately involved in the key metal chelation and oxygen activation event required for DNA cleavage.[1] Thus, **2–4** constitute important substructures of the pyrimidoblamic acid subunit accessible only through chemical synthesis. These agents along with (−)-desmethylpyrimidoblamic acid, *epi*-(+)-desmethylpyrimidoblamic acid, and (+)-desmethyldesacetamidopyrimidoblamic acid disclosed in recent efforts[13] provide a key

Scheme 1



series of structurally modified pyrimidoblamic acid analogs. Their incorporation into a full range of structural analogs of deglycobleomycin A_2 is detailed in the accompanying article.[16]

The approach to the authentic and modified pyrimidoblamic acid subunits is based on the inverse electron demand Diels–Alder reaction[17] of 2,4,6-tris(ethoxycarbonyl)-1,3,5-triazine (**5**)[13,18,19] for the preparation of the pyrimidine nucleus central to the structure of the agents. Key to the completion of the synthesis of (−)-pyrimidoblamic acid was the development and implementation of a diastereoselective imine addition reaction of optically active *N*-acyloxazolidinones for the stereocontrolled introduction of the C2 acetamido side chain.[20] Chemical studies which establish and confirm the absolute configuration of the C2 acetamido side chain are detailed, and their extension to the diastereoselective synthesis of **4** is described.

1,3,5-Triazine → Pyrimidine Heteroaromatic Diels–Alder Reaction: Synthesis of the Pyrimidine Core. Two concise approaches to the preparation of **9** based on the [4 + 2] cycloaddition of 2,4,6-tris(ethoxycarbonyl)-1,3,5-triazine (**5**) have been developed (Scheme 1). Treatment of **5**, prepared in one step by the acid-catalyzed trimerization of ethyl cyanoformate (95–100%),[18] with 1-(dibenzylamino)propyne (**6**,[21] 2 equiv) provided **8** in excellent yield (95–98%) under thermal reaction conditions (101 °C, dioxane, 21 h). The room temperature [4 + 2] cycloaddition reaction of **5** with **6** is followed by a subsequent retro Diels–Alder reaction with loss of ethyl cyanoformate, and it is the rate of the cycloreversion reaction that dictates the required

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Table 1. Representative Results of a Study of the [4 + 2] Cycloaddition Reactions of **5** with **6** and **7**

entry	conditions	product	yield (%)
1	DMF, 90 °C, 29 h	**9**	75
2	DMF, 100 °C, 72 h	**9**	80
3	DMF, 110 °C, 22 h	**9**	65
4	DMF, 120 °C, 48 h	**9**	53
5	DMF, 130 °C, 29 h	**9**	46
6	DMF, 140 °C, 26 h	**9**	44
7	DMF, K_2CO_3, 100 °C, 48 h	**9**	45
8	$PhCH_3$, CH_3CO_2H, 100 °C, 72 h	**9**	7
9	dioxane, 101 °C, 48 h	**9**	0
10	dioxane, 101 °C, 21 h	**8**	95
11	CH_3CN, 82 °C, 12 h	**8**	73

thermal reaction conditions. Acid-catalyzed debenzylation of **8** under vigorous conditions (CF_3SO_3H, CH_2Cl_2, 40 °C, 13 h, 75%) provided **9**, and initial efforts to effect the conversion of **8** to **9** through catalytic hydrogenolysis proved less successful. In general, the deprotection of **8** under a range of alternative conditions gave predominantly the monodebenzylation product in addition to recovered starting material. Alternatively, **9** was derived directly and conveniently in one step by treatment of **5** with propionamidine hydrochloride (**7**, 100 °C, DMF, 72 h, 80%) in a reaction cascade that proceeds with thermal tautomerization of **7** to 1,1-diaminopropene and its [4 + 2] cycloaddition reaction with **5**. The sequential elimination of ammonia, imine to enamine tautomerization, and subsequent retro Diels–Alder loss of ethyl cyanoformate under the reaction conditions provided **9** directly in excellent yield. The use of a polar aprotic solvent, the use of the hydrochloride salt of **7**, and the carefully defined thermal conditions facilitate both the amidine tautomerization and aromatization of the initial cycloadduct (Table 1). Notably, the use of lower reaction temperatures (<80 °C) provided lower conversions to **9** presumably due to inadequate amidine to enamine tautomerization. Higher reaction temperatures (>100 °C) similarly provided lower conversions to **9** and presumably may be attributed to competitive reactions of the substrates, **9**, and the reaction cascade intermediates under the more vigorous reaction conditions. The use of the propionamidine free base resulted in lower yields of **9** (45%, DMF, 100 °C, 48 h), and efforts to employ the corresponding methyl imidate or imidate hydrochloride proved significantly less successful.

Synthesis of (+)-Desacetamidopyrimidoblamic Acid (3). Key to the use of **9** in the synthesis of **1–4** was the selective differentiation of the pyrimidine C2 and C4 ethyl esters. This transformation was accomplished through selective reduction of the sterically and electronically more accessible C2 ethoxycarbonyl group of **9** to provide **10**. Characteristic of the enhanced electrophilic nature of the C2 ethoxycarbonyl group responsible for the selective reduction, the reaction was effectively conducted with sodium borohydride at low temperature (1.0 equiv, EtOH, 5 °C, 150 h, 70%; Scheme 2). In a study of the reduction of **9**, it was observed that the selectivity of the competitive C2 and C4 ethoxycarbonyl reductions improved as the reactivity of the reagent was reduced and the reaction temperature was lowered (Table 2). A number of more reactive reducing agents conventionally employed to reduce esters failed to provide the required C2 versus C4 reduction selectivity including $LiBH_4$, DIBAL-H, and L- and K-Selectride. 2D ^1H–^1H NOESY NMR of the isomeric alcohols unambiguously confirmed the isomer assignments through observation of a diagnostic CH_2OH/C5-CH_3 NOE crosspeak for the minor isomer and its absence with **10**. Alternative efforts to differentiate the C2 and C4 ethyl esters through selective acid- or base-catalyzed hydrolysis, transesterification, or aminolysis proved less successful and less direct than the selective $NaBH_4$ reduction of **9**.

Conversion of **10** to the tosylate **11** (99%), which proceeded cleanly without competitive sulfonamide generation, followed by

Scheme 2

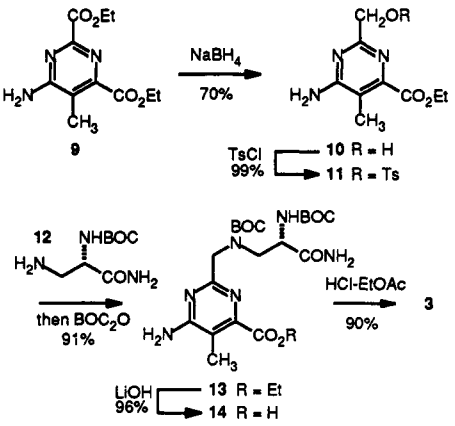

Table 2. Representative Results of the Selective Reduction of **9**

entry	conditions[a]	2:4-CH_2OH	yield (%)
1	*t*BuOH–EtOH (5:1), 80 °C, 1 h	2:1	30
2	EtOH, 25 °C, 15 h	4:1	49
3	EtOH, 10 °C, 108 h	23:1	51
4	EtOH, 5 °C, 150 h	28:1	70
5	MeOH, –25 °C, 72 h	no reaction	0
6	Me_4NBH_4, EtOH, 25 °C, 96 h	17:1	36
7	$Zn(BH_4)_2$, THF–Et_2O (1:1), 25 °C	6:1	10

[a] $NaBH_4$ unless indicated otherwise.

clean displacement with **12**[13] (4 equiv, 2 equiv of $NaHCO_3$, CH_3CN, 25 °C, 10 h) and subsequent protection of the secondary amine provided **13** (91% overall for two steps). Hydrolysis of the ethyl ester (LiOH, 96–100%) provided the N^{α},N^{β}-bis((*tert*-butyloxy)carbonyl) derivative of desacetamidopyrimidoblamic acid (**14**), $[\alpha]^{22}_D$ –9.2 (*c* 0.25, CH_3OH) (lit[4a] $[\alpha]^{25}_D$ –8.9 (*c* 1.25, CH_3OH)), and subsequent acid-catalyzed deprotection (3 N HCl–EtOAc, 25 °C, 30 min, 90%) of **14** provided (+)-desacetamidopyrimidoblamic acid (**3**), $[\alpha]^{22}_D$ +8.1 (*c* 0.15, 0.1N HCl).

Diastereoselective *N*-Acyloxazolidinone Enolate–Imine Addition Reaction: Preparation of (–)-Pyrimidoblamic Acid (1) and *epi*-(+)-Pyrimidoblamic Acid (2). The final strategic element necessary for completion of the synthesis of pyrimidoblamic acid was the stereocontrolled introduction of the C2 acetamido side chain. Prior studies have relied on nonselective chemical approaches requiring a separation of the resulting 1:1 mixture of diastereomers.[4] In parallel with studies on the total synthesis of P-3A,[13] we have examined the potential diastereoselective addition of optically active enolates with imines and found that the use of the Evans' *N*-acyloxazolidinones[22] provided a diastereoselective imine addition reaction suitable for the C2 acetamido side chain introduction.[20]

Oxidation of **10** (10 equiv of MnO_2, CH_3CN, 82 °C, 3 h, 83%) followed by condensation of **15** with **12**[13] (98–100%) afforded **16** (Scheme 3). The modest conversions observed under standard MnO_2 oxidation conditions (10 equiv, CH_2Cl_2, 25 °C, 25–40%) that may be attributed principally to the limited solubility of **10** and **15** were improved upon conducting the reaction in refluxing acetonitrile. Further use of dilute reaction conditions (0.05 M) eliminated a minor competitive self condensation reaction with imine formation and provided excellent conversions of **10** to **15**. A range of alternative oxidants including PCC, PDC, $Ba(MnO_4)_2$ (CH_2Cl_2, 25 °C, 25%), and TFAA–DMSO were not as successful at providing **15** due to the solubility properties of the substrate under conventional reaction conditions.

Addition of the stannous (*Z*)-enolate **17**, generated by treatment

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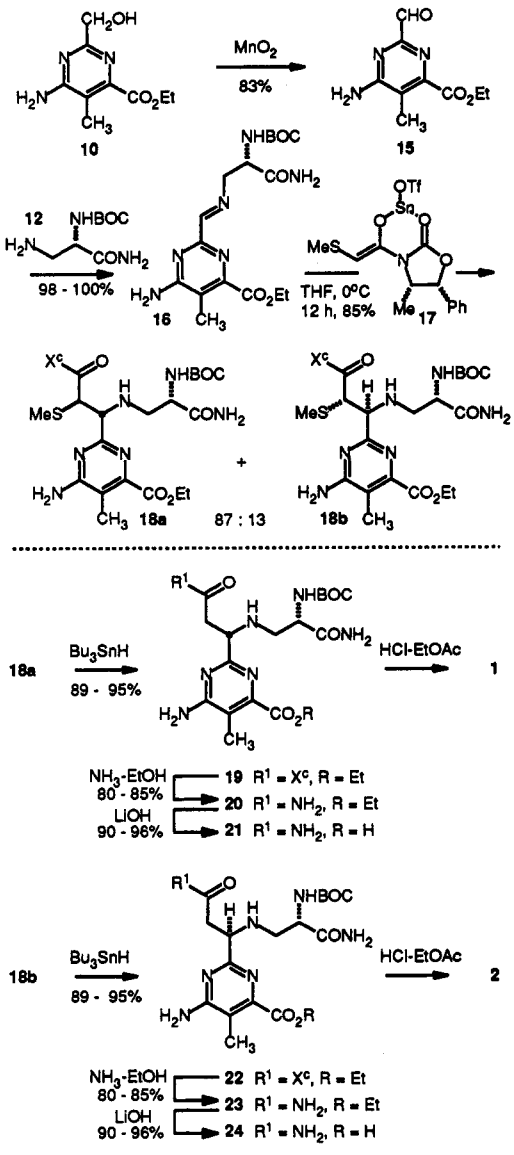
Scheme 3



of the corresponding oxazolidinone[23] (1 equiv, THF, –20 °C, 1 h) with iPr$_2$NEt (2.2 equiv) in the presence of Sn(OTf)$_2$ (2.0 equiv), to **16** (0.5 equiv) provided a separable 87:13 mixture of the imine addition adducts **18a** and **18b** (THF, 0 °C, 12 h, 81–85%). The prescribed reaction conditions were derived only through considerable experimentation and ultimately provided the opportunity to use a minimal number of protecting groups for the potentially reactive functionality found in imine **16**. Key to the success of the diastereoselective imine addition reaction[24,25] was the use of the stannous enolate[25c,26,27] (2.0 equiv) in the presence of two additional equivalents of Sn(OTf)$_2$, and under such conditions the major *anti*-**18a** adduct was found to slowly epimerize to *syn*-**18a** (0 °C, 12 h, 16:1 *anti*:*syn*-**18a** versus 0 °C, 24 h, 1.8:1 *anti*:*syn*-**18a**). The stannous enolate **17** proved much more effective than the titanium enolate[28,29] (20–30% yield, 9:1 **18a** and **18b**), which provided the same products with a comparable level of diastereoselection but in much lower conversions, and the corresponding di-*n*-butylboronyl enolate of **17** proved ineffective. The stannous enolate of (4*S*,5*R*)-3-acetyl-4-methyl-5-phenyl-2-oxazolidinone provided a 1:1 mixture of **19** and **22** (56%), indicating an important role for the thiomethyl group of **17** in the diastereoselection of the imine addition reaction. The analogous stannous enolate of (4*S*,5*R*)-2-((phenylthio)acetyl)-4-methyl-5-phenyl-2-oxazolidinone provided the imine adducts with a comparable diastereoselectivity (9:1, 72%) but in slightly lower chemical conversion, and the adduct diastereomers were not readily separable. Reductive desulfurization of the major diastereomer, *anti*-**18a** as well as *syn*-**18a** (Bu$_3$SnH, 89–95%), followed by aminolysis of **19** (16% NH$_3$–EtOH, 0 °C, 1 h, 80–85%) provided **20**; $[\alpha]^{25}_D$ –10.8 (*c* 0.36, EtOH) (lit[4a] $[\alpha]^{25}_D$ –7.5 (*c* 1.0, EtOH)). Longer reaction periods for the reaction with NH$_3$ led to subsequent aminolysis of the ethyl ester. Hydrolysis of the ethyl ester **20** (LiOH, 90–95%) provided the well-characterized N^{α}-((*tert*-butyloxy)carbonyl) derivative of (–)-pyrimidoblamic acid **21**, mp 220–222 °C, $[\alpha]^{25}_D$ –35.6 (*c* 0.8, H$_2$O) (lit[4a] mp 220–222 °C, $[\alpha]^{28}_D$ –33.6 and –32.8 (*c* 0.75, H$_2$O)). Acid-catalyzed deprotection (3 N HCl–EtOAc, 25 °C, 1 h, 100%) of **21** provided (–)-pyrimidoblamic acid (**1**), $[\alpha]^{25}_D$ –27 (*c* 0.12, H$_2$O). Subjection of the minor diastereomer **18b** (>20:1 *anti*:*syn*) to the identical sequence provided **22–24**, mp 221–223 °C, $[\alpha]^{25}_D$ +20.8 (*c* 0.44, H$_2$O) (lit[4a] mp 221 °C, $[\alpha]^{25}_D$ +20.8 (*c* 0.65, H$_2$O)) and *epi*-(+)-pyrimidoblamic acid (**2**), $[\alpha]^{25}_D$ +20.1 (*c* 0.11, H$_2$O).

The diastereoselection observed in the imine addition reaction which we have investigated in some detail deserves further discussion. Unlike the boron enolate, which might be anticipated to react as a nonchelated enolate through a closed-chair transition state to provide the Evans' syn addition product,[22,23] the expanded coordination sphere of tin(II) like that of titanium(IV)[28,29] was expected to permit reaction of the chelated enolate **17** with **16** with the additional complexation and activation of the imine to provide the non-Evans' syn imine addition product. However, the reaction provided not the chelated enolate syn imine addition product but rather the corresponding chelated enolate anti adduct as the major product. In retrospect, this potentially may be attributed to reaction of the imine in its preferred *E* configuration with both large imine substituents occupying axial positions in a closed-chair transition state further reinforced through additional internal chelation of the imine pyrimidinyl nitrogen with tin(II).[27] Although it is not possible to rule out reaction of the chelated (*Z*)-enolate **17** with imine **16** activated by external coordination to the added Lewis acid and proceeding through an open transition state,[30] the lack of reaction of the corresponding di-*n*-butylboronyl (*Z*)-enolate with **16** in the presence of added Lewis acid catalysts (Sn(OTf)$_2$, Et$_2$AlCl) suggests this may prove unlikely.

The assignment of the relative and absolute stereochemistry of the imine addition products was made based on the following observations. Reductive desulfurization of *anti*-**18a** and *syn*-**18a** both provided **19**, which was found to possess the natural *S* configuration at the newly introduced amine center through conversion to natural (–)-pyrimidoblamic acid (**1**). In addition, deliberate epimerization of the major addition product (*anti*-**18a**) provided *syn*-**18a**, isomeric at the thiomethyl center. In

(23) Evans, D. A.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2127.

(24) Hart, D. J.; Ha, D.-C. *Chem. Rev.* **1989**, *89*, 1447. Brown, M. J. *Heterocycles* **1989**, *29*, 2225.

(25) (a) Corey, E. J.; Decicco, C. P.; Newbold, R. C. *Tetrahedron Lett.* **1991**, *32*, 5287. (b) Ojima, I.; Habus, I. *Tetrahedron Lett.* **1990**, *31*, 4289. (c) Nagao, Y.; Dai, W.-M.; Ochiai, M.; Tsukagoshi, S.; Fujita, E. *J. Org. Chem.* **1990**, *55*, 1148. Nagao, Y.; Dai, W.-M.; Ochiai, M. *Tetrahedron Lett.* **1988**, *29*, 6133. (d) Hart, D. J.; Lee, C.-S.; Pirkle, W. H.; Hyon, M. H.; Tsipouras, A. *J. Am. Chem. Soc.* **1986**, *108*, 6054. (e) Yamada, T.; Suzuki, H.; Mukaiyama, T. *Chem. Lett.* **1987**, 293, **1986**, 915. Yamasaki, N.; Murakami, M.; Mukaiyama, T. *Chem. Lett.* **1986**, 1013. (f) Furukawa, M.; Okawara, T.; Noguchi, Y.; Terawaki, Y. *Chem. Pharm. Bull.* **1978**, *26*, 260. (g) Shibasaki, M.; Ishida, Y.; Iwasaki, G.; Iimori, T. *J. Org. Chem.* **1987**, *52*, 3488. (h) Georg, G. I.; Kant, J.; Gill, H. S. *J. Am. Chem. Soc.* **1987**, *109*, 1129.

(26) Evans, D. A.; Weber, A. E. *J. Am. Chem. Soc.* **1986**, *108*, 6757. Evans, D. A.; Gage, J. R.; Leighton, J. L.; Kim, A. S. *J. Org. Chem.* **1992**, *57*, 1961.

(27) Kobayashi, S.; Hachiya, I. *J. Org. Chem.* **1992**, *57*, 1324.

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(29) Evans, D. A.; Clark, J. S.; Metternich, R.; Novack, V. J.; Sheppard, G. S. *J. Am. Chem. Soc.* **1990**, *112*, 866.

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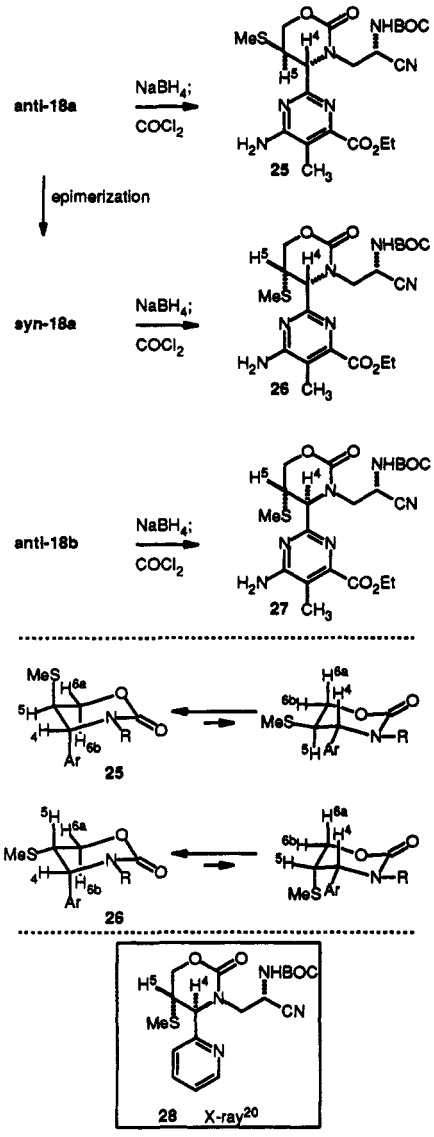
Scheme 4



contrast, desulfurization of *anti*-**18b** provided **22**, which proved to be a diastereomer of **19**. The relative stereochemistry of *anti*-**18a**, *syn*-**18a**, and *anti*-**18b** was established upon conversion to the cyclic carbamates **25**–**27** (Scheme 4).

For the cyclic carbamate **25** derived from the major addition product, the C4-H/C5-H coupling constant was determined to be 1.5 Hz, diagnostic of an equatorial–equatorial H-5/H-4 relationship, and proved analogous to that of **28** for which a single-crystal X-ray structure had unambiguously established the trans stereochemistry.[20] Notably, the cyclic carbamate **25** adopts a conformation in which both the thiomethyl and aryl substituents occupy axial positions, and is the result of the lack of 1,3-diaxial MeS/H interactions and the presence of a single 1,3-diaxial Ar/H interaction within the preferred diaxial conformation. The alternative diequatorial conformation suffers from two significant and destabilizing gauche interactions of the vicinal ring substituents. Similarly, carbamate **27** derived from the minor imine addition product exhibits a C4-H/C5-H coupling constant of 1.2 Hz, characteristic of the trans stereochemistry, and, like **25**, is derived from an imine addition product possessing the anti stereochemistry. In contrast, carbamate **26** derived from *syn*-**18a** exhibited a characteristic C4-H/C5-H coupling constant of 5.6 Hz, diagnostic of an axial–equatorial H-4/H-5 relationship in which H-5 occupies an axial position. Consistent with these assignments, the C5-H $w_{1/2}$values of 6.8 Hz (**25** and **27**) and 19.4 Hz (**26**) were observed and proved diagnostic of an equatorial and axial C5-H, respectively. Further consistent with the assignments, **25** and **27** exhibited ^{1}H–^{1}H NOE crosspeaks in the 2D-NOESY NMR for MeS/H-6a, H-5, and H-4. Notably, crosspeaks for MeS/H-6b and H-4/H-6a were not observed and would be diagnostic of an equatorial versus axial thiomethyl substituent and a 1,3-diaxial H-4/H-6a relationship, respectively. Carbamate **26** exhibited ^{1}H–^{1}H NOE crosspeaks in the 2D-NOESY NMR for MeS/H-6a, H-6b, H-4, and H-5 as well as a diagnostic H-6b/$CO_2CH_2CH_3$ crosspeak, and no H-4/H-6a crosspeak was observed. The former are consistent with an equatorial versus axial thiomethyl substituent, and the latter two observations are diagnostic of a conformation in which the aryl group occupies an axial position. Finally, calculated coupling constants for H-4/H-5, H-5/H-6a, and H-5/H-6b for the analogous MM2 low-energy conformations of **25** or **27** (obsd 1.5, 1.5, and 2.0 Hz; calcd 1.4, 2.6, and 1.4 Hz) and for **26** (obsd 5.6, 4.9, 8.2 Hz; calcd 4.3, 5.2, 11.7 Hz) agree with the observed coupling constants of the structural assignments.

Confirmation of the Absolute Stereochemistry. The absolute configuration assigned to the pyrimidoblamic acid and bleomycin A_2 center which bears the C2 acetamido side chain was established in a single-crystal X-ray structure determination of **29**[31,32] derived from a chemical degradation product obtained under vigorous acidic conditions (6 N HCl, 105 °C, 20 h) resulting in partial (15–30%) epimerization of the amine center under consideration.[33,34] This partial epimerization of the precursor to **29** coupled with the use of a nonheavy-atom derivative in the crystallographic solution of its absolute configuration has resulted in some concern as to the accuracy of the assigned absolute stereochemistry. This proved to be of special concern since additional spectroscopic properties of **29** provided a tentative assignment of the opposite absolute configuration.[31] Unfortunately, the existing syntheses of **1** which provided a 1:1 mixture of isomers at the critical amine center have relied on correlation with the natural product to provide their stereochemical assignments and thus have not addressed the confirmation of the pyrimidoblamic acid assigned stereochemistry. In the course of our work, this uncertainty had become an independent concern in the ongoing NMR studies in which the preliminary assignment of the NOEs of bleomycin A_2 metal complexes was not easily reconciled on the basis of assigned absolute stereochemistry.[35] This long-standing ambiguity in the absolute stereochemistry of **1** as well as the inability of our own asymmetric synthesis of pyrimidoblamic acid to permit unambiguous assignment of absolute configuration at the C2 acetamido side chain center provided the incentive for us to establish it by an additional chemical means.

Chemical degradation of naturally derived bleomycin A_2[36] under strong acidic conditions (6 N aqueous HCl, 105 °C, 20 h) followed by immediate esterification (3 N HCl, CH_3OH, reflux, 1 h) of the dicarboxylic acid **30**, which itself proved difficult to isolate, provided **31**, $[\alpha]^{25}_{365}$ +26 (*c* 0.15, 1 N HCl). In turn, acid-catalyzed hydrolysis of the readily purified diester **31** (3 N aqueous HCl, 100 °C, 1 h) provided **30**, $[\alpha]^{25}_{365}$ +30 (*c* 0.2, 1 N HCl) (lit[31] $[\alpha]^{20}_{365}$ +47.5 (*c* 0.2, 1 N HCl) (Scheme 5). Exposure of **20** to the same set of reaction conditions similarly provided **31** (75%), $[\alpha]^{25}_{365}$ +37 (*c* 0.15, 1 N HCl), $[\alpha]^{25}_{D}$ +3.4 (*c* 0.15, 1 N HCl), and **30** (94%), $[\alpha]^{25}_{365}$ +40 (*c* 0.07, 1 N HCl), confirming the correlation of **18a**, **19**–**21**, and **1** with naturally derived material. In contrast, exposure of **23** to the same set of

(31) Nakamura, H.; Yoshioka, T.; Takita, T.; Umezawa, H.; Muraoka, Y.; Iitaka, Y. *J. Antibiot.* **1976**, *29*, 762.
(32) Iitaka, Y.; Nakamura, H.; Nakatani, T.; Muraoka, Y.; Fujii, A.; Takita, T.; Umezawa, H. *J. Antibiot.* **1978**, *31*, 1070.
(33) Takita, T.; Muraoka, Y.; Maeda, K.; Umezawa, H. *J. Antibiot.* **1968**, *21*, 79.
(34) Muraoka, Y.; Takita, T.; Maeda, K.; Umezawa, H. *J. Antibiot.* **1970**, *23*, 252.
(35) For example, see: Marzilli, L. G.; Freyder, C. P. *Magn. Reson. Chem.* **1991**, *29*, 338.
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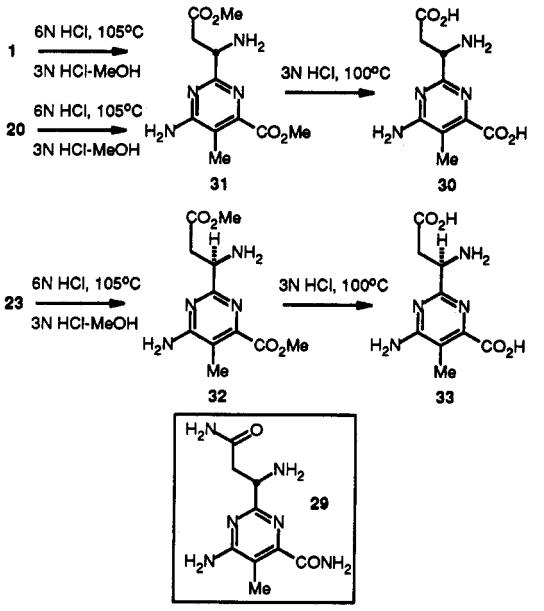
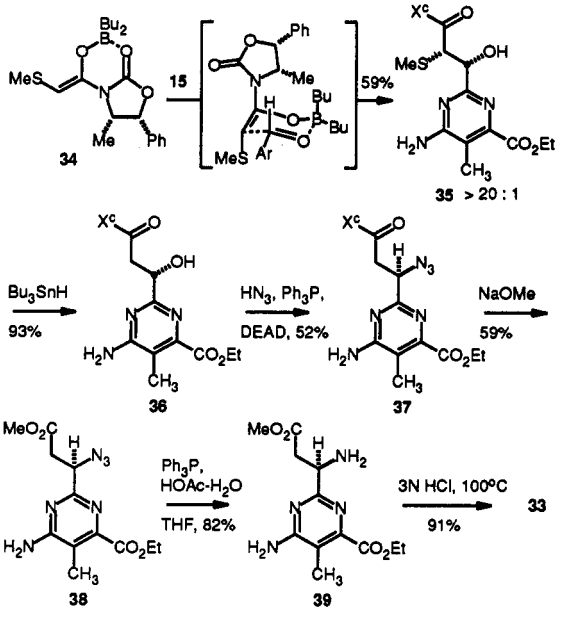
Scheme 5



Scheme 6

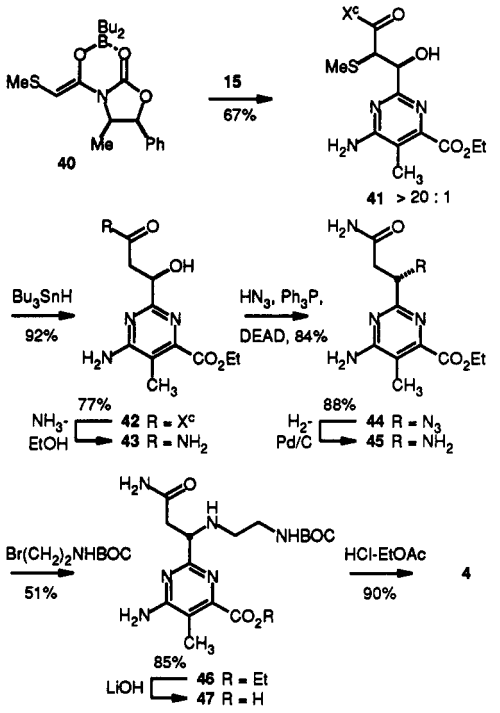


reaction conditions provided **32** (64%), $[\alpha]^{25}_{365}$ −39 (*c* 0.14, 1 N HCl), $[\alpha]^{25}_{D}$ −3.7 (*c* 0.14, 1 N HCl), and **33** (94%), $[\alpha]^{25}_{365}$ −40 (*c* 0.08, 1 N HCl), enantiomeric with **31**–**30** and confirming the diastereomeric stereochemical assignments for **18b**, **22**–**24**, and **2**.

With this set of authentic correlation samples in hand, we elected to prepare **30**–**31** and **32**–**33** by asymmetric synthesis to permit the unambiguous assignment of stereochemistry. Treatment of aldehyde **15** with the di-*n*-butylboronyl (*Z*)-enolate **34**[23] provided the Evans' (2*S*,3*R*)-*syn*-aldol adduct **35** as the only detectable product (59%, >20:1 syn) in which the resident chirality on the optically active oxazolidinone dictates the relative and absolute stereochemistry at the newly introduced centers with reaction of the nonchelated boron enolate through a closed-chair transition state (Scheme 6). Bu_3SnH reductive cleavage of the thiomethyl substituent followed by direct azide displacement[37] of the alcohol upon Mitsunobu activation[38] provided **37** with clean

(37) Loibner, H.; Zbiral, E. *Helv. Chim. Acta* **1977**, *60*, 417.
(38) Mitsunobu, O. *Synthesis* **1981**, 1.

Scheme 7



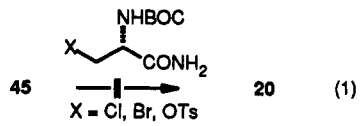
inversion of the stereochemistry.[39] Removal of the optically active oxazolidinone through methanolysis followed by reduction of the azide to the corresponding amine provided diester **39**, $[\alpha]^{25}_{365}$ −54 (*c* 0.16, 1 N HCl), $[\alpha]^{25}_{D}$ −5.6 (*c* 0.16, 1 N HCl).[40] Acid-catalyzed hydrolysis of **39** (3 N aqueous HCl, 100 °C, 3 h) provided diacid **33**, $[\alpha]^{25}_{365}$ −59 (*c* 0.11, 1 N HCl), enantiomeric with naturally derived material. Thus, consistent with the original absolute stereochemical assignments, **30**, **31**, pyrimidoblamic acid, and the bleomycins possess the *S* configuration as shown. The use of the enolate **40**, enantiomeric with **34**, provided comparable intermediates possessing the natural *S* configuration as detailed in the following efforts.

Preparation of (−)-Descarboxamidopyrimidoblamic Acid (4) and Exploration of an Alternative Diastereospecific Synthesis of (−)-Pyrimidoblamic Acid (1). Given the enhanced diastereoselection achieved with use of the aldol versus imine addition reaction of the *N*-acyloxazolidinone enolates, we elected to investigate the potential of its implemention in an alternative synthesis of (−)-pyrimidoblamic acid (**1**). Further adaptation of this approach was anticipated to provide a synthesis of **4** through the divergent and late stage introduction of a modified C2 side chain not as easily accessible through use of our initial strategy. Treatment of **15** with the di-*n*-butylboronyl (*Z*)-enolate **40**[23] provided (2*R*,3*S*)-*syn*-**41** as the only detectable reaction product (67%, >20:1) (Scheme 7). Reductive desulfurization of **41** effected by treatment with Bu_3SnH for removal of the thiomethyl group provided **42** (93%). Aminolysis of the *N*-acyloxazolidinone conducted at 0 °C (77%) followed by Mitsunobu activation of the alcohol **43** and azide displacement[37] with inversion of the stereochemistry provided **44** in excellent yield (84%) with no evidence of loss of stereochemical integrity at the reaction center and with observance of only a trace amount of the corresponding elimination product. The reverse order of reactions with Mitsunobu activation and azide displacement[37] of the alcohol **42**

(39) Some elimination (*ca.* 15–20%) accompanies the displacement reaction on **36**, but no isomeric azide was detected. In the sequence leading to **4**, this competitive reaction was reduced to a trace side reaction by conducting the displacement on the carboxamide **43**.
(40) Efforts to convert the oxazolidinone to the methyl ester prior to azide introduction led to competitive elimination versus azide introduction, and efforts to reduce the azide prior to oxazolidinone methanolysis led to competitive hydrolysis of the oxazolidinone.

followed by aminolysis of the *N*-acyloxazolidinone also provided **44** but in lower conversions (51 × 40%). Reduction of the azide to the corresponding amine **45** was accomplished best by mild catalytic hydrogenolysis (H_2, Pd-C, CH_3OH, 25°C, 1 h, 88%), but **45** was also obtained in good yield upon treatment of **44** with Ph_3P (H_2O–HOAc, THF, 25 °C, 16 h, 71%). Alkylation of **45** with *N*-BOC-2-bromoethylamine (K_2CO_3, DMF, 0.4 M, 25 °C, 48 h) provided **46**. Alternative efforts to conduct the alkylation reaction with the corresponding tosylate (DMF, CH_3CN, EtOH, acetone; 2–4 days, 25 °C, 18–30%) provided lower conversions with recovered **45**, and efforts to drive the reaction to completion using more vigorous reaction conditions led to competitive amine elimination (DMF, 50 °C) or dialkylation (DMF, 6–13 kbar, 25 °C). Hydrolysis (LiOH) of the ethyl ester followed by acid-catalyzed deprotection of **47**, $[\alpha]^{25}_D$ –8.0 (*c* 0.2, H_2O), provided (–)-descarboxamidopyrimidoblamic acid (**4**), $[\alpha]^{25}_D$ –18 (*c* 0.05, H_2O).

Efforts to extend this approach to an alternative synthesis of (–)-pyrimidoblamic acid itself have not yet proven successful (eq 1). Significant, although not exhaustive, efforts to alkylate **45**



(1)

with β-chloro-, β-bromo-, or β-tosyl-*N*-BOC-L-alanine amide under a wide variety of conditions provided little or no **20** and generally provided recovered **45** and products derived from decomposition (elimination) of the alkylating agent. Alternative efforts to activate the alcohol of **43** toward direct displacement with β-amino-*N*-BOC-L-alanine amide by formation of the corresponding tosylate (21%) suffered from competitive elimination.

The incorporation of **1–4** into the total synthesis of bleomycin A_2 and structurally related agents is detailed in the accompanying articles.

Experimental Section

2,4-Bis(ethoxycarbonyl)-6-(dibenzylamino)-5-methylpyrimidine (8). A solution of 2,4,6-tris(ethoxycarbonyl)-1,3,5-triazine[18] (**5**, 4.11 mmol, 1.22 g) in dioxane (10 mL) under Ar was treated with 1-(dibenzylamino)-1-propyne[21] (**6**, 8.22 mmol, 1.93 g, 2.0 equiv) at 25 °C, and the resulting reaction solution was warmed at 101 °C (21 h). Removal of the solvent in vacuo and flash chromatography (SiO_2, 4 × 15 cm, 20–40% EtOAc–hexane gradient elution) afforded pure **8** (1.69 g, 1.78 g theoretical, 95%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.32 (10H, m), 4.73 (4H, s, 2 × CH_2Ph), 4.45 (2H, q, *J* = 7.2 Hz, CH_2CH_3), 4.44 (2H, q, *J* = 7.2 Hz, CH_2CH_3), 2.36 (3H, s, CH_3), 1.42 (3H, t, *J* = 7.2 Hz, CH_2CH_3), 1.39 (3H, t, *J* = 7.2 Hz, CH_2CH_3); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 165.8 (e, C-6), 165.7 (e, CO_2Et), 163.7 (e, CO_2Et), 157.9 (e, C-2), 153.2 (e, C-4), 136.9 (e), 128.5 (o), 127.9 (o) and 127.4 (o, four C_6H_5), 116.3 (e, C-5), 62.1 (e, two CH_2CH_3), 52.4 (e, two CH_2Ph), 16.0 (o, CH_3), 14.1 (o, CH_2CH_3), 14.0 (o, CH_2CH_3); IR (film) ν_{max} 3029, 2982, 2935, 1739, 1560, 1496, 1453, 1427, 1242, 1222, 1091, 800 cm^{-1}; UV ($CHCl_3$) λ_{max} 278 (ε 13 400), 244 (ε 6700) nm; CIMS (2-methylpropane) *m/e* 434 (M^+ + H, base); CIHRMS (2-methylpropane) *m/e* 434.2054 (M^+ + H, $C_{25}H_{27}N_3O_4$ requires 434.2080).

Anal. Calcd for $C_{25}H_{27}N_3O_4$: C, 69.28; H, 6.24; N, 9.70. Found: C, 69.30; H, 6,29; N, 9.45.

6-Amino-2,4-bis(ethoxycarbonyl)-5-methylpyrimidine (9). Method A: A solution of **8** (1.42 mmol, 616 mg) in anhydrous CH_2Cl_2 (7 mL) was treated with CF_3SO_3H (14.2 mmol, 2.13 g, 10 equiv) at 25 °C, and the resulting reaction mixture was warmed at 40 °C (13 h). The mixture was cooled to 0 °C, diluted with H_2O (10 mL), and treated with 2 N aqueous NaOH (7.5 mL). Additional 2 N aqueous NaOH was added to further adjust the pH to 9, the mixture was extracted with CH_2Cl_2 (3 × 20 mL), and the combined organic phase was dried ($MgSO_4$). Removal of the solvent under reduced pressure and flash chromatography (SiO_2, 3 × 15 cm, 60% EtOAc–hexane) afforded pure **9** (270 mg, 359 mg theoretical, 75%) as a white solid identical to that described below.

Method B: A solution of 2,4,6-tris(ethoxycarbonyl)-1,3,5-triazine[18] (**5**, 9.0 mmol, 2.69 g) in anhydrous DMF (45 mL) under Ar was treated with propionamidine hydrochloride (**7**, 18.0 mmol, 1.92 g, 2.0 equiv) at 25 °C, and the resulting reaction mixture was warmed at 100 °C (72 h). Removal of the solvent in vacuo and recrystallization (EtOAc–hexane) afforded pure **9** (1.82 g, 2.28 g theoretical, 80%) as a white solid: mp 155–156 °C (white needles, EtOAc–hexane); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 6.22 (2H, s, NH_2), 4.47 (2H, q, *J* = 7.1 Hz, CH_2CH_3), 4.45 (2H, q, *J* = 7.1 Hz, CH_2CH_3), 2.30 (3H, s, CH_3), 1.44 (3H, t, *J* = 7.1 Hz, CH_2CH_3), 1.43 (3H, t, *J* = 7.1 Hz, CH_2CH_3); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 165.3 (e, C-6), 164.1 (e, C_4-CO_2Et), 163.7 (e, C_2-CO_2Et), 157.9 (e, C-2), 153.6 (e, C-4), 114.8 (e, C-5), 62.5 (e, CH_2CH_3), 62.2 (e, CH_2CH_3), 14.1 (o, CH_2CH_3), 14.0 (o, CH_2CH_3), 12.2 (o, CH_3); UV ($CHCl_3$) λ_{max} 294 (ε 3900), 252 (ε 7100), 236 (ε 2700) nm; IR (KBr) ν_{max} 3448, 3360, 2980, 2938, 1734, 1720, 1628, 1576 cm^{-1}; EIMS *m/e* (relative intensity) 253 (M^+, 1), 181 (100), 135 (19), 107 (16), 81 (21); CIMS (2-methylpropane) *m/e* (relative intensity) 254 (M^+ + H, base); EIHRMS *m/e* 253.1060 (M^+, $C_{11}H_{15}N_3O_4$ requires 253.1062).

Anal. Calcd for $C_{11}H_{15}N_3O_4$: C, 52.12; H, 5.92; N, 16.58. Found: C, 51.91; H, 6.11; N, 16.34.

6-Amino-4-(ethoxycarbonyl)-2-(hydroxymethyl)-5-methylpyrimidine (10). A solution of **9** (2.0 mmol, 506 mg) in anhydrous EtOH (10 mL) was cooled to 5 °C and treated with $NaBH_4$ (2.0 mmol, 75.7 mg, 1.0 equiv) under Ar. After being stirred at 5 °C for 150 h, the reaction mixture was treated with saturated aqueous $NaHCO_3$ (10 mL) and aqueous H_2O_2 (5%, 10 mL) at 0 °C and was stirred for 5 h. The mixture was extracted with 20% 2-propanol–$CHCl_3$ (5 × 20 mL), and the combined organic extracts were dried ($MgSO_4$). Removal of solvent in vacuo afforded a mixture of the 2- and 4-(hydroxymethyl)pyrimidines (2-CH_2OH:4-CH_2OH 28:1) as a white solid. The two isomeric alcohols were separated by preparative centrifugal thin-layer chromatography (SiO_2, 5% DMF–EtOAc) to afford pure **10** (312 mg, 422 mg theoretical, 70%) as a white solid: mp 169 °C sharp (white needles, EtOAc–hexane); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.77 (2H, br s, NH_2), 4.63 (2H, s, CH_2-OH), 4.43 (2H, q, *J* = 7.1 Hz, CH_2CH_3), 3.55 (1H, br s, OH), 2.22 (3H, s, CH_3), 1.41 (3H, t, *J* = 7.1 Hz, CH_2CH_3); 2D ^{1}H–^{1}H NOESY NMR ($CDCl_3$, 200 MHz) did not reveal an NOE crosspeak between CH_2OH and C5-CH_3 for **10** but did so for the minor isomer derived from the $NaBH_4$ reduction of **9**; ^{13}C NMR (DMSO-d_6, 50 MHz) δ 166.9 (e, C=O), 166.5 (e, C-2), 164.1 (e, C-6), 154.2 (e, C-4), 108.7 (e, C-5), 64.6 (e, CH_2OH), 61.5 (e, CH_2CH_3), 14.2 (o, CH_2CH_3), 11.9 (o, CH_3); IR (KBr) ν_{max} 3396, 3340, 3168, 2986, 2936, 1722, 1664, 1542, 1508 cm^{-1}; EIMS *m/e* (relative intensity) 211 (M^+, 2), 139 (89), 110 (13), 108 (15), 85 (13), 81 (100); CIMS (2-methylpropane) *m/e* (relative intensity) 212 (M^+ + H, base); EIHRMS *m/e* 211.0956 (M^+, $C_9H_{13}N_3O_3$ requires 211.0956).

Anal. Calcd for $C_9H_{13}N_3O_3$: C, 51.18; H, 6.16; N, 19.91. Found: C, 50.93, H, 6.39; N, 19.62.

6-Amino-4-(ethoxycarbonyl)-5-methyl-2-(((*p*-tolylsulfonyl)oxy)methyl)pyrimidine (11). A solution of **10** (2.56 mmol, 540 mg) in CH_2Cl_2 (6 mL) was treated with K_2CO_3 (5.12 mmol, 707 mg, 2.0 equiv) and *p*-TsCl (2.56 mmol, 488 mg, 1.0 equiv) at 25 °C under Ar, and the resulting reaction mixture was stirred at 25 °C for 20 h. The crude reaction mixture was filtered through a Celite pad (CH_2Cl_2, 3 × 10 mL), and the solvent was removed in vacuo. Chromatography (SiO_2, 3 × 7 cm, 50% EtOAc–hexane) afforded pure **11** (1.00 g, 1.01 g theoretical, 99%) as a white solid: mp 128 °C sharp (white needles, EtOAc–hexane); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.84 (2H, d, *J* = 8.1 Hz, $SO_2C_6H_4CH_3$), 7.33 (2H, d, *J* = 8.3 Hz, $SO_2C_6H_4CH_3$), 6.31 (2H, br s, NH_2), 5.01 (2H, s, CH_2-OTs), 4.41 (2H, q, *J* = 7.2 Hz, CH_2CH_3), 2.44 (3H, s, $SO_2C_6H_4CH_3$), 2.22 (3H, s, CH_3), 1.40 (3H, t, *J* = 7.2 Hz, CH_2CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 165.7 (e, C=O), 164.4 (e, C-2), 159.1 (e, C-6), 152.8 (e, C-4), 145.5 (e), 131.6 (e), 130.0 (o), 128.1 (o), 112.0 (e, C-5), 70.7 (e, CH_2), 62.0 (e, CH_2CH_3), 21.6 (o, $SO_2C_6H_4CH_3$), 14.1 (o, CH_2CH_3), 11.7 (o, CH_3); IR (KBr) ν_{max} 3457, 3328, 3181, 2980, 1720, 1654, 1570, 1559 cm^{-1}; CIMS (2-methylpropane) *m/e* (relative intensity) 366 (M^+ + H, 18), 230 (base); FABHRMS (NBA) *m/e* 366.1124 (M^+ + H, $C_{16}H_{19}N_3O_5S$ requires 366.1124).

Anal. Calcd for $C_{16}H_{19}N_3O_5S$: C, 52.60; H, 5.21; N, 11.51; S, 8.77. Found: C, 52.84; H, 5.37; N, 11.14; S, 8.69.

N^{α},N^{β}-Bis((*tert*-butyloxy)carbonyl)-N^{β}-[(6-amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)methyl]-(*S*)-β-aminoalanine Amide (13). A solution of **11** (0.63 mmol, 248 mg) and **12**[13] (2.5 mmol, 508 mg, 4.0 equiv) in CH_3CN (5.0 mL) was treated with $NaHCO_3$ (1.25 mmol, 105 mg, 2 equiv), and the mixture was stirred under Ar at 25 °C for 10 h. Removal of solvent in vacuo afforded the crude product, which was dissolved in

THF-saturated aqueous $NaHCO_3$ (1:1, 5 mL) and treated with di-*tert*-butyl dicarbonate (2.5 mmol, 550 mg, 0.57 mL, 4 equiv), and the resulting mixture was stirred at 25 °C for 8 h. Water (5 mL) was added, and the mixture was extracted with 20% 2-propanol–$CHCl_3$ (3 × 20 mL). The combined organic extracts were dried ($MgSO_4$), and the solvent was removed in vacuo. Chromatography (SiO_2, 1 × 3 cm, 50–100% EtOAc–hexane gradient elution) afforded **13**[41] (276 mg, 310 mg theoretical, 89%; typically 89–91% for two steps) as a white solid: mp 174 °C sharp (EtOAc–hexane); $[\alpha]^{22}_D$ +27.9 (*c* 0.19, $CHCl_3$); ^1H NMR (DMSO-d_6, 200 MHz) δ 7.50–6.70 (5H, m, NH_2, $CONH_2$, NHBOC), 4.40-4.10 (3H, m, CH_2, CH_2CH), 4.30 (2H, q, J = 7.2 Hz, CH_2CH_3), 3.62 (1H, dd, J = 5.7, 13.9 Hz, $CHHCH$), 3.45–3.25 (1H, m, $CHHCH$), 2.00 (3H, s, CH_3), 1.45–1.20 (21H, m, CH_2CH_3, 2 × $C(CH_3)_3$); ^1H NMR (acetone-d_6, 200 MHz) δ 7.80–6.40 (5H, m), 4.80–4.10 (3H, m), 4.35 (2H, q, J = 7.0 Hz), 3.90 (1H, dd, J = 6.0, 14.0 Hz), 3.60–3.40 (1H, m), 2.15 (3H, s), 1.45–1.20 (21H, m); ^{13}C NMR (DMSO-d_6, 50 MHz) δ 172.9 (e, C=O), 166.7 (e, C=O), 164.7 (e, C=O), 164.3 (e, C=O), 164.0 (e, C-2), 155.8 (e, C-6), 154.1 (e, C-4), 108.4 (o, C-5), 79.4 (e, $C(CH_3)_3$), 79.0 (e, $C(CH_3)_3$), 78.5 (e, CH_2), 78.4 (e, CH_2CH), 61.4 (e, CH_2CH_3), 53.3 (o, CH_2CH), 28.3 (o, CH_3), 28.0 (o, CH_2CH_3), 14.0 (o, $C(CH_3)_3$), 11.8 (o, $C(CH_3)_3$); IR (KBr) ν_{max} 3358, 2978, 2934, 1686, 1650, 1582 cm^{-1}; CIMS (2-methylpropane) *m/e* (relative intensity) 497 (M^+ + H, base); CIHRMS (2-methylpropane) *m/e* 497.2714 (M^+ + H, $C_{22}H_{36}N_6O_7$ requires 497.2724).

Anal. Calcd for $C_{22}H_{36}N_6O_7$: C, 53.23; H, 7.26; N, 16.94. Found: C, 53.07; H, 7.30; N, 16.71.

N^{α},N^{β}-Bis((*tert*-butyloxy)carbonyl)-N^{β}-[(6-amino-4-carboxypyrimidin-2-yl)methyl]-(*S*)-β-aminoalanine Amide (14). A solution of **13** (0.093 mmol, 46 mg) in THF–CH_3OH–H_2O (3:1:1, 0.5 mL) at 25°C was treated with 1 N aqueous LiOH (0.19 mmol, 0.19 mL, 2 equiv), and the mixture was stirred for 4 h. After evaporation of most of the THF–CH_3OH, the aqueous phase was extracted with $CHCl_3$ (2 × 2 mL). The aqueous phase was acidified to pH 4–5 with the addition of 1.2 N aqueous HCl and was extracted with 20% 2-propanol–$CHCl_3$ (5 × 10 mL). The combined organic extracts were dried ($MgSO_4$), and the solvent was removed in vacuo to afford **14** (42 mg, 44 mg theoretical, 96%) as a white foam: $[\alpha]^{22}_D$ –9.2 (*c* 0.25, CH_3OH) (lit[44a] $[\alpha]^{25}_D$ –8.9 (*c* 1.25, CH_3OH)); R_f 0.18 (SiO_2, 7:3 $CHCl_3$–CH_3OH); ^1H NMR (D_2O, 400 MHz) δ 4.33–4.39 (3H, m, CH_2NBOC, CH_2CH), 3.5-3.7 (2H, m, CH_2CH), 2.09 (3H, br s, CH_3), 1.27–1.13 (18H, m, 2 × $C(CH_3)_3$); IR (CH_3OH) ν_{max} 3506, 3412, 3130, 2978, 2896, 1695, 1684, 1519, 1472, 1425, 1360, 1249, 1161, 1043, 732, cm^{-1}; FABHRMS (NBA) *m/e* 469.2420 (M^+ + H, $C_{20}H_{32}N_6O_7$ requires 469.2411).

(+)-Desacetamidopyrimidoblamic Acid (3). A solution of **14** (0.021 mmol, 9.9 mg) in EtOAc (0.1 mL) was cooled to 0 °C and treated with 3 N HCl in EtOAc (3 mL). The resulting reaction mixture was stirred at 25 °C for 30 min. Removal of solvent in vacuo afforded **3** as a white solid. Pure **3** (7.1 mg, 7.9 mg theoretical, 90%) was obtained by trituration with anhydrous CH_2Cl_2 (3 × 5 mL) and Et_2O (3 × 5 mL): $[\alpha]^{22}_D$ +1.8 (*c* 0.55, CH_3OH), $[\alpha]^{22}_D$ +8.1 (*c* 0.15, 0.1 N HCl); ^1H NMR (CD_3OD, 400 MHz) δ 4.47 (1H, m, CH_2CH), 4.41 (2H, br s, CH_2), 3.65 (2H, m, CH_2CH), 2.41 (3H, br s, CH_3); IR (KBr) ν_{max} 3628, 3417, 3347, 3006, 1716, 1690, 1684, 1652, 1636, 1576, 1506, 1457, 1395, 1109, 1090 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 401.0338 (M^+ + Cs, $C_{10}H_{16}N_6O_3$ requires 401.0342).

6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidine-2-carboxaldehyde (15). A solution of **10** (0.3 mmol, 71 mg) in anhydrous CH_3CN (6.7 mL) was treated with activated MnO_2 (3.36 mmol, 293 mg, 10 equiv) at 25 °C, and the resulting suspension was warmed at 82 °C for 3 h. The cooled reaction mixture was filtered through a Celite pad (CH_3CN, 5 × 10 mL), and the solvent was removed in vacuo. Flash chromatography (SiO_2, 0.5 × 3 cm, 80% EtOAc–hexane) afforded pure **15** (58 mg, 70 mg theoretical, 83%) as a white foam: ^1H NMR ($CDCl_3$, 300 MHz) δ 9.93 (1H, s, CHO), 5.75 (2H, br s, NH_2), 4.49 (2H, q, J = 7.1 Hz, OCH_2CH_3), 2.33 (3H, s, CH_3), 1.45 (3H, t, J = 7.1 Hz, OCH_2CH_3); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 192.2 (o, CHO), 165.6 (e, CO_2Et), 164.5 (e, C-2), 157.0 (e, C-6), 153.9 (e, C-4), 116.2 (e, C-5), 62.4 (e, CH_2CH_3), 29.5 (o, CH_3), 13.9 (o, CH_2CH_3); IR (KBr) ν_{max} 3351, 3009, 2973, 2932, 2902, 1731, 1696, 1656, 1621 cm^{-1}; EIMS *m/e* (relative intensity) 209 (M^+, 4), 180 (5), 137 (57), 109 (16), 92 (20), 81 (82), 69 (35), 57 (base); CIMS (2-methylpropane) *m/e* (relative intensity) 210 (M^+ + H, base); EIHRMS *m/e* 209.0800 (M^+, $C_9H_{11}N_3O_3$ requires 209.0800).

Anal. Calcd for $C_9H_{11}N_3O_3$: C, 51.65; H, 5.30; N, 20.10. Found: C, 51.60; H, 5.48; N, 20.05.

N^{α}-((*tert*-Butyloxy)carbonyl)-N^{β}-[[(6-amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)methylene]amino]-(*S*)-β-aminoalanine Amide (16). A solution of **15** (0.215 mmol, 45 mg) and **12**[13] (0.215 mmol, 44 mg, 1.0 equiv) in anhydrous CH_3CN (4 mL) was treated with 4-Å molecular sieves (500 mg) at 25 °C. The resulting suspension was stirred at 25 °C for 24 h. The reaction mixture was filtered through a Celite pad (CH_2Cl_2, 5 × 5 mL). Removal of solvent in vacuo afforded **16** (83 mg, 84.7 mg theoretical, 98%) as a white foam: $[\alpha]^{25}_D$ +92.1 (*c* 0.45, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 8.28 (1H, s, CH=N), 7.06 (2H, m), 5.71 (1H, d, J = 8.0 Hz, HNBOC), 4.58 (1H, m, CH_2CH), 4.45 (2H, q, J = 7.1 Hz, CH_2CH_3), 4.11 (1H, dd, J = 3.3, 13.1 Hz, $CHHCH$), 3.93 (1H, dd, J = 5.6, 13.1 Hz, $CHHCH$), 2.28 (3H, s, CH_3), 1.41 (12H, m, CH_3, $C(CH_3)_3$); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 174.1 (e, C=N), 173.4 (e, C=O), 165.8 (e, C=O), 164.2 (e, C-2), 164.0 (e, C-6), 158.0 (e, C=O), 152.8 (e, C-4), 114.9 (e, C-5), 80.0 (e, $C(CH_3)_3$), 62.4 (e, CH_2CH_3), 62.0 (o, CH_2CH), 55.3 (e, CH_2CH), 28.2 (o, CH_3), 28.1 (o, $C(CH_3)_3$), 14.0 (o, CH_2CH_3); IR ($CHCl_3$) ν_{max} 3619, 3415, 3010, 2978, 1725, 1687, 1577, 1501, 1422, 1215, 1072, 1047, 928 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 527.1019 (M^+ + Cs, $C_{17}H_{26}N_6O_5$ requires 527.0977).

Diastereoselective Reaction of the Stannous (*Z*)-Enolate of (4*S*,5*R*)-3-((Methylthio)acetyl)-4-methyl-5-phenyl-2-oxazolidinone with 16. Tin(II) trifluoromethanesulfonate (1.6 mmol, 663 mg, 4.0 equiv) was dissolved in dry THF (2 mL) under N_2, cooled to –78 °C, and treated sequentially with (4*S*,5*R*)-3-((methylthio)acetyl)-4-methyl-5-phenyl-2-oxazolidinone[23] (0.78 mmol, 211 mg, 2.0 equiv) in dry THF (2 mL) and i-Pr_2NEt (1.75 mmol, 226 mg, 0.31 mL, 4.4 equiv). The mixture was stirred for 1 h at –20 °C for complete enolization, and the reaction mixture was recooled to –78 °C. A solution of **16** (0.40 mmol, 157 mg) in dry THF (2 mL) was added, and the reaction mixture was allowed to warm to 0 °C, where it was stirred for 12 h. The reaction mixture was poured into a two-layer solution of CH_2Cl_2 (30 mL) and saturated aqueous $NaHCO_3$ (15 mL) with vigorous stirring. The organic layer was washed with saturated aqueous NaCl (2 × 7 mL), dried ($MgSO_4$), and concentrated in vacuo. Chromatography (SiO_2, 3 × 6 cm, 5% CH_3OH–CH_2Cl_2) gave 213 mg (262 mg theoretical, 81%) of a 82:5:13 mixture of three diastereomers (87:13 **18a** and **18b**) determined to be the anti product (*anti*-**18a**), the corresponding syn product (*syn*-**18a**), and third diastereomer, *anti*-**18b**, possessing the undesired 3*R* configuration. Upon prolonged reaction times, the ratio of the three products changed (24 h, 0 °C, 46:39:15), indicating the in situ epimerization of the C2 thiomethyl center, but the critical **18a**:**18b** ratio remained unchanged (85–87:15–13). Preparative HPLC separation (SiO_2, 97:3 $CHCl_3$–CH_3OH) of the mixture of products afforded *anti*-**18a** (160 mg), *syn*-**18a** (10 mg), and the third diastereomer (*anti*-**18b**, 24 mg) as white solids.

Ethyl 2(*R*)-[1-[[2(*S*)-[((*tert*-Butyloxy)carbonyl)amino]-2-carbamoylethyl]amino]-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]-2(*R*)-(methylthio)ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (*anti*-18a). Mp 124–127 °C (EtOAc–hexane); R_f 0.34 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ –27.8 (*c* 0.44, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.50–7.20 (5H, m), 6.97 (2H, br s), 5.81 (2H, br s), 5.75 (1H, d, J = 7.2 Hz), 5.74 (1H, d, J = 8.2 Hz), 4.94 (1H, d, J = 11.2 Hz), 4.84 (1H, dq, J = 6.5, 7.2 Hz), 4.44 (2H, q, J = 7.1 Hz), 4.23 (1H, d, J = 11.2 Hz), 3.98 (1H, br s), 3.08 (1H, dd, J = 3.1, 13.1 Hz), 2.62 (1H, br s), 2.25 (3H, s), 2.14 (3H, s), 1.41 (3H, t, J = 7.1 Hz), 1.36 (9H, s), 0.99 (3H, d, J = 6.5 Hz); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 174.1, 169.5, 166.0, 165.2, 164.3, 155.7, 153.0, 152.7, 132.7, 128.6, 128.5, 125.5, 111.2, 79.3, 78.7, 64.1, 61.5, 55.0, 53.2, 49.2, 46.2, 27.9, 14.6, 13.9, 11.8, 11.3; IR ($CHCl_3$) ν_{max} 3416, 2985, 1777, 1725, 1689, 1612, 1570, 1363, 1223, 1196, 1120, 1070 cm^{-1}; FABHRMS (NBA) *m/e* 660.2820 (M^+ + H, $C_{30}H_{41}N_7O_8S$ requires 660.2816).

Anal. Calcd for $C_{30}H_{41}N_7O_8S$: C, 54.59; H, 6.26; N, 14.87. Found: C, 54.39; H, 6.14; N, 14.52.

Ethyl 2(*R*)-[1-[[2(*S*)-[((*tert*-Butyloxy)carbonyl)amino]-2-carbamoylethyl]amino]-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]-2(*S*)-(methylthio)ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (*syn*-18a). Mp 123–135 °C (EtOAc–hexane); R_f 0.30 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ –6.5 (*c* 0.39, $CHCl_3$); ^1H NMR (CD_3OD, 400 MHz) δ 7.50–7.20 (5H, m), 5.69 (1H, d, J = 8.1 Hz), 5.29 (1H, d, J = 10.6 Hz), 4.65 (1H, dq, J = 6.5, 7.1 Hz), 4.35 (2H, q, J = 7.1 Hz), 2.88 (1H, dd, J = 6.7, 13.0 Hz), 2.80 (1H, dd, J = 5.6, 12.7 Hz), 2.11 (3H, s), 2.10 (3H, s), 1.41 (9H, s), 1.34 (3H, t, J = 7.1 Hz), 0.77 (3H, d, J = 6.5 Hz); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 173.8, 169.7, 165.7, 165.6, 163.4, 155.6,

(41) The amine displacement of the 2-(chloromethyl)pyrimidine provided **13** in a comparable conversion. In addition, **13** was prepared from imine **16** through catalytic hydrogenation (0.1 wt equiv of PtO_2, H_2, 25 °C, 16 h) and subsequent protection of the secondary amine (BOC_2O), but the sequence detailed in Scheme 2 proved superior.

152.1, 151.8, 132.9, 132.6, 128.2, 125.2, 110.3, 78.4, 78.2, 61.4, 60.5, 54.0, 53.2, 47.7, 46.1, 27.9, 14.3, 13.8, 11.4, 11.3; IR ($CHCl_3$) ν_{max} 3515, 3413, 1778, 1687, 1612, 1570, 1450, 1368, 1223, 1199, 1075 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 792.1800 (M^+ + Cs, $C_{30}H_{41}N_7O_8S$ requires 792.1792).

Ethyl 2(*S*)-[1-[[2(*S*)-[((*tert*-Butyloxy)carbonyl)amino]-2-carbamoylethyl]amino]-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]-2(*S*)-(methylthio)ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (*anti*-18b). Mp 111–114 °C (EtOAc–hexane); R_f 0.32 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +14.8 (*c* 0.27, $CHCl_3$); ^1H NMR (CD_3OD, 400 MHz) δ 7.50–7.25 (5H, m), 5.87 (1H, d, *J* = 8.1 Hz), 5.05 (1H, d, *J* = 10.6 Hz), 4.94 (1H, dq, *J* = 6.5, 7.2 Hz), 4.42 (2H, q, *J* = 7.2 Hz), 4.15 (1H, d, *J* = 10.6 Hz), 4.04 (1H, m), 2.87 (1H, dd, *J* = 3.5, 13.2 Hz), 2.75 (1H, dd, *J* = 6.0, 13.1 Hz), 2.17 (3H, s), 2.02 (3H, s), 1.42 (3H, t, *J* = 7.2 Hz), 1.41 (9H, s), 0.81 (3H, d, *J* = 6.5 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.6, 171.0, 167.7, 167.5, 165.8, 157.5, 154.3, 153.8, 135.2, 129.8, 129.5, 127.0, 112.0, 80.5, 80.2, 65.4, 63.0, 55.7, 55.3, 51.0, 48.5, 28.7, 14.6, 14.5, 12.2, 12.0; IR ($CHCl_3$) ν_{max} 3403, 2993, 1778, 1723, 1687, 1612, 1573, 1452, 1364, 1221, 1198, 1071 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 792.1760 (M^+ + Cs, $C_{30}H_{41}N_7O_8S$ requires 792.1792).

Ethyl 2(*S*)-[1-[[2(*S*)-[((*tert*-Butyloxy)carbonyl)amino]-2-carbamoylethyl]amino]-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]-ethyl]-6-amino-5-methylpyrimidine- 4-carboxylate (19). A solution of *anti*-**18a** (0.29 mmol, 192 mg) in C_6H_6 (2 mL) was treated with Bu_3SnH (0.87 mmol, 250 mg, 3.0 equiv) and AIBN (0.042 mmol, 7 mg, 0.15 equiv) and was warmed at 80 °C (45 min) under N_2. The mixture was allowed to cool to 23 °C, and the solvent was evaporated in vacuo. Flash chromatography (SiO_2, 3 × 10 cm, 5% CH_3OH–CH_2Cl_2) afforded **19** as a white solid (162 mg, 178 mg theoretical, 91%): mp 122–124 °C (EtOAc–hexane); R_f 0.28 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ −19.3 (*c* 0.29, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.75 (1H, br s), 7.43 (1H, m), 7.40–7.20 (5H, m), 6.01 (1H, br s), 5.74 (2H, br s), 5.67 (1H, d, *J* = 7.2 Hz), 5.58 (1H, br s), 5.47 (1H, m), 4.76 (1H, dq, *J* = 6.6, 7.2 Hz), 4.41 (2H, q, *J* = 7.1 Hz), 4.27 (1H, m), 4.02 (1H, m), 3.47 (1H, dd, *J* = 8.4, 16.0 Hz), 3.28 (1H, d, *J* = 14.8 Hz), 3.15 (1H, d, *J* = 11.8 Hz), 2.62 (1H, dd, *J* = 7.0, 13.1 Hz), 2.19 (3H, s), 1.43 (9H, s), 1.40 (3H, t, *J* = 7.1 Hz), 0.90 (3H, d, *J* = 6.6 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.2, 172.0, 168.8, 167.4, 165.6, 157.5, 154.5, 154.2, 135.0, 129.4, 129.2, 126.9, 111.1, 80.5, 80.1, 62.8, 61.0, 55.7, 55.2, 49.7, 41.4, 28.4, 14.7, 14.2, 11.8; IR ($CHCl_3$) ν_{max} 3415, 3010, 1780, 1689, 1611, 1569, 1493, 1370, 1223, 1197, 1069 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 746.1914 (M^+ + Cs, $C_{29}H_{39}N_7O_8$ requires 746.1914).

Anal. Calcd for $C_{29}H_{39}N_7O_8$: C, 56.74; H, 6.41; N, 15.98. Found: C, 56.72; H, 6.31; N, 15.47.

Similar treatment of *syn*-**18a** provided **19**, identical in all respects.

N^{α}-((*tert*-Butyloxy)carbonyl)-N^{β}-[1-amino-3(*S*)-(6-amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)propion-3-yl]-(*S*)-β-aminoalanine Amide (20). Solid **19** (0.16 mmol, 99 mg) was treated with an ethanolic solution of NH_3 (16%, 20 mL), and the solution was stirred for 1.5 h at 0 °C. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 3 × 5 cm, 10% CH_3OH–CH_2Cl_2) afforded **20** as a white solid (59 mg, 73 mg theoretical, 80%): mp 157–159 °C (*i*PrOH-hexane) (lit[4a] mp 159–162 °C (*i*PrOH)); R_f 0.32 (20% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ −10.8 (*c* 0.36, EtOH) (lit[4a] $[\alpha]^{25}_D$ −7.5 (*c* 1.0, EtOH)); ^1H NMR (CD_3OD, 400 MHz) δ 4.31 (2H, q, *J* = 7.1 Hz), 4.03 (1H, m), 3.88 (1H, dd, *J* = 4.9, 8.8 Hz), 2.69 (2H, m), 2.53 (1H, dd, *J* = 5.0, 15.0 Hz), 2.45 (1H, dd, *J* = 8.9, 15.0 Hz), 2.04 (3H, s), 1.35 (9H, s), 1.29 (3H, t, *J* = 7.1 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.5, 176.4, 168.3, 167.7, 165.9, 157.8, 154.4, 111.5, 80.8, 63.0, 62.0, 55.7, 50.0, 42.0, 28.7, 14.5, 12.1; IR (neat) ν_{max} 3416, 2978, 1727, 1678, 1611, 1517, 1423, 1214, 1046, 929 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 586.1396 (M^+ + Cs, $C_{19}H_{31}N_7O_6$ requires 586.1390).

N^{α}-((*tert*-Butyloxy)carbonyl)-N^{β}-[1-amino-3(*S*)-(6-amino-4-carboxy-5-methylpyrimidin-2-yl)propion-3-yl]-(*S*)-β-aminoalanine Amide (21). A solution of **20** (0.046 mmol, 20.7 mg) in THF–CH_3OH–H_2O (3:1:1, 0.5 mL) at 0 °C was treated with aqueous 1 N LiOH (0.07 mmol, 0.07 mL, 1.5 equiv), and the mixture was stirred for 1.5 h. After evaporation of most of the THF–CH_3OH, the aqueous phase was acidified to pH 4–5 with the addition of aqueous 1.2 N HCl and the solvent was evaporated in vacuo. The residue was charged onto a column of Dowex (1 × 8 cm, acetate form, 50–100 mesh). The column was washed with H_2O, and subsequent elution with 6% HOAc–H_2O afforded **21** as a white powder (17.6 mg, 19 mg theoretical, 91%): mp 220–222 °C (EtOH-hexane) (lit[4a] mp 220–222 °C (EtOH)); R_f 0.58 (4:1:1 *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ −35.6 (*c* 0.81, H_2O) (lit $[\alpha]^{28}_D$ −33.6[4a] and −32.8[4b] (*c* 0.75, H_2O)); ^1H NMR (D_2O, 400 MHz) δ 4.17 (2H, m), 3.16 (1H, m), 3.03 (1H, m), 2.76 (2H, m), 1.93 (3H, s), 1.30 (9H, s); ^{13}C NMR (CD_3OD, 100 MHz) δ 174.6, 169.0, 166.5, 161.1, 157.6, 154.2, 112.9, 81.6, 60.6, 52.1, 48.5, 36.3, 28.6, 12.2; IR (neat) ν_{max} 3423, 3189, 1720, 1685, 1479, 1258, 1161, 1022, 878 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 426.2118 (M^+ + H, $C_{17}H_{27}N_7O_6$ requires 426.2101).

(−)-Pyrimidoblamic Acid (1). The solid **21** (0.005 mmol, 2.1 mg) was treated with 3 N HCl–EtOAc (0.5 mL), and the mixture was stirred at 25 °C for 1 h. The solvent was evaporated in vacuo to give pure **1** hydrochloride (2.2 mg, 2.2 mg theoretical, 100%) as a clear, hygroscopic solid: R_f 0.32 (4:1:1 *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ −26.7 (*c* 0.12, H_2O); ^1H NMR (D_2O, 400 MHz) δ 4.24 (1H, dd, *J* = 7.9, 7.9 Hz), 4.12 (1H, dd, *J* = 4.5, 6.6 Hz), 3.21 (1H, dd, *J* = 4.5, 13.7 Hz), 3.08 (1H, dd, *J* = 6.6, 13.7 Hz), 2.89 (1H, dd, *J* = 5.4, 15.8 Hz), 2.82 (1H, dd, *J* = 7.4, 15.8 Hz), 2.22 (3H, s); IR (neat) ν_{max} 3456, 3247, 1695, 1681, 1557, 1161, 1078, 820 cm^{-1}; FABHRMS (NBA) *m/e* 326.1564 (M^+ + H, $C_{12}H_{19}N_7O_4$ requires 326.1577).

Ethyl 2(*R*)-[1-[[2(*S*)-[((*tert*-Butyloxy)carbonyl)amino]-2-carbamoylethyl]amino]-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]-ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (22). A solution of **18b** (0.09 mmol, 58 mg) in C_6H_6 (2 mL) was treated with Bu_3SnH (0.26 mmol, 77 mg, 3 equiv) and AIBN (6.02 mmol, 3 mg, 0.15 equiv), and the solution was warmed at 80 °C (45 min) under N_2. The mixture was allowed to cool to 23 °C, and the solvent was evaporated in vacuo. Flash chromatography (SiO_2, 3 × 3 cm, 5% CH_3OH–CH_2Cl_2) afforded **22** (49 mg, 53 mg theoretical, 92%) as a white solid: mp 121–123 °C (EtOAc–hexane); R_f 0.28 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ −1.1 (*c* 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.40–7.20 (6H, m), 6.40 (1H, m), 5.79 (2H, br s), 5.71 (1H, d, *J* = 7.2 Hz), 5.50 (2H, m), 4.77 (1H, dq, *J* = 6.6, 7.2 Hz), 4.42 (2H, q, *J* = 7.2 Hz), 4.35 (1H, dd, *J* = 5.5, 7.5 Hz), 4.11 (1H, m), 3.39 (2H, m), 3.16 (1H, m), 2.71 (1H, dd, *J* = 6.5, 13.0 Hz), 2.18 (3H, s), 1.45 (9H, s), 1.40 (3H, t, *J* = 7.2 Hz), 0.91 (3H, d, *J* = 6.6 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.4, 172.2, 168.1, 167.5, 165.6, 157.5, 154.6, 154.3, 135.1, 129.5, 129.3, 126.9, 111.1, 80.5, 80.1, 62.8, 60.9, 55.7, 55.5, 49.8, 41.6, 32.5, 14.6, 14.3, 11.9; IR ($CHCl_3$) ν_{max} 3418, 3018, 2981, 1729, 1687, 1611, 1573, 1455, 1368, 1215, 1122, 1069, 780 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 746.1944 (M^+ + Cs, $C_{29}H_{39}N_7O_8$ requires 746.1914).

N^{α}-((*tert*-Butyloxy)carbonyl)-N^{β}-[1-amino-3(*R*)-(6-amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)propion-3-yl]-(*S*)-β-aminoalanine Amide (23). Compound **22** (0.064 mmol, 39 mg) was subjected to aminolysis as described for **19** to provide **23** (24 mg, 29 mg theoretical, 82%) as a white solid: mp 69–72 °C (*i*PrOH–hexane) (lit[4a] mp 64–66 °C (*i*PrOH)); R_f 0.32 (20% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ +38 (*c* 0.57, EtOH) (lit[4a] $[\alpha]^{25}_D$ +14.8 (*c* 1.0, EtOH)); ^1H NMR (CD_3OD, 400 MHz) δ 4.31 (2H, q, *J* = 7.1 Hz), 4.01 (1H, m), 3.88 (1H, dd, *J* = 5.3, 8.3 Hz), 2.75 (1H, dd, *J* = 5.3, 12.4 Hz), 2.68 (1H, dd, *J* = 6.4, 10.4 Hz), 2.47 (2H, m), 2.04 (3H, s), 1.33 (9H, s), 1.29 (3H, t, *J* = 7.1 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.7, 168.4, 167.8, 165.9, 157.8, 154.4, 111.4, 80.7, 63.0, 61.9, 55.8, 50.0, 41.8, 28.7, 14.5, 12.1; IR (neat) ν_{max} 3407, 2996, 1725, 1653, 1609, 1519, 1474, 1420, 1215, 1047, 927, 880 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 586.1396 (M^+ + Cs, $C_{19}H_{31}N_7O_6$ requires 586.1390).

N^{α}-((*tert*-Butyloxy)carbonyl)-N^{β}-[1-amino-3(*R*)-(6-amino-4-carboxy-5-methylpyrimidin-2-yl)propion-3-yl]-(*S*)-β-aminoalanine Amide (24). Compound **23** (0.022 mmol, 9.9 mg) was subjected to hydrolysis and purification as described for **20** to provide **24** (8.8 mg, 9.3 mg theoretical, 94%): mp 221–223 °C (EtOH–hexane) (lit[4a] mp 221 °C); R_f 0.58 (4:1:1, *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ +20.8 (*c* 0.44, H_2O) (lit[4a] $[\alpha]^{25}_D$ +20.8 (*c* 0.65, H_2O)); ^1H NMR (D_2O, 400 MHz) δ 4.08 (2H, m), 3.06 (2H, m), 2.76 (2H, m), 2.00 (3H, s), 1.30 (9H, s); ^{13}C NMR (CD_3OD, 100 MHz) 174.5, 174.3, 168.4, 166.5, 160.9, 157.6, 152.9, 113.9, 81.7, 61.0, 52.0, 48.5, 35.7, 28.6, 12.2; IR (neat) ν_{max} 3412, 3145, 1717, 1686, 1584, 1479, 1370, 1258, 1160, 873 cm^{-1}; FABHRMS (NBA) *m/e* 426.2118 (M^+ + H, $C_{17}H_{27}N_7O_6$ requires 426.2101).

***epi*-(+)-Pyrimidoblamic Acid (2).** The solid **24** (0.005 mmol, 2.2 mg) was subjected to acid-catalyzed deprotection in the same fashion as **21** to give *epi*-(+)-pyrimidoblamic acid hydrochloride (**2**, 2.4 mg, 2.4 mg theoretical, 100%) as a clear, hygroscopic solid: R_f 0.32 (4:1:1 *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ +20.1 (*c* 0.11, H_2O); ^1H NMR (D_2O, 400 MHz) δ 4.25 (1H, dd, *J* = 6.6, 6.6 Hz), 4.10 (1H, dd, *J* = 5.8, 5.8 Hz), 3.18 (1H, dd, *J* = 5.4, 13.5 Hz), 3.12 (1H, dd, *J* = 6.4, 13.6 Hz), 2.90 (1H, dd, *J* = 5.8, 16.6 Hz), 2.82 (1H, dd, *J* = 7.1, 16.0 Hz), 2.21 (3H, s); IR (neat) ν_{max} 3448, 3256, 1692, 1682, 1468, 1161, 1021, 810 cm^{-1}; FABHRMS (NBA) *m/e* 326.1567 (M^+ + H, $C_{12}H_{19}N_7O_4$ requires 326.1577).

Carbamate 25.[42] A solution of *anti*-**18a** (52 mg, 0.079 mmol) in anhydrous CH_3OH (1 mL) was cooled to 0 °C and treated with $NaBH_4$ (20 mg, 0.55 mmol, 7 equiv) under Ar. After being stirred at 0 °C for 1 h, the reaction mixture was quenched with the addition of H_2O (0.2 mL). The mixture was extracted with 10% CH_3OH–CH_2Cl_2 (2 × 5 mL), and the combined organic extracts were dried (Na_2SO_4) and concentrated in vacuo. Flash chromatography (SiO_2, 3 × 5 cm, 5% CH_3OH–CH_2Cl_2) gave the corresponding alcohol as a white foam. A solution of the alcohol (0.011 mmol, 4.5 mg) in anhydrous CH_2Cl_2 (0.2 mL) was treated with pyridine (20 mg, 0.25 mmol) and $COCl_2$ (0.087 mmol, 45 μL, 1.93 M in toluene) at 25 °C. After being stirred at 25 °C for 30 min, the reaction mixture was poured into a two-layer solution of CH_2Cl_2 and saturated aqueous $NaHCO_3$. The organic layer was washed with 10% aqueous HCl, saturated aqueous $NaHCO_3$, and saturated aqueous NaCl, dried (Na_2SO_4), and concentrated in vacuo. Flash chromatography (SiO_2, 3 × 5 cm, 5% CH_3OH–CH_2Cl_2) gave **25** (1.7 mg, 4.3 mg theoretical, 40%) as a white foam: R_f 0.33 (10% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ +14.5 (*c* 0.06, $CHCl_3$); ^1H NMR (CD_3OD, 400 MHz) δ 4.78 (1H, m), 4.59 (1H, d, *J* = 1.5 Hz, C4-H), 4.47 (1H, dd, *J* = 2.0, 11.0 Hz), 4.30 (2H, q, *J* = 7.2 Hz), 4.08 (1H, dd, *J* = 1.5, 11.0 Hz), 3.75 (1H, dd, *J* = 7.3, 13.5 Hz), 3.38 (1H, br s), 3.21 (1H, m), 2.18 (3H, s), 2.05 (3H, s), 1.33 (9H, s), 1.29 (3H, t, *J* = 7.2 Hz); IR ($CHCl_3$) ν_{max} 3010, 2976, 2399 (s, C≡N), 1734, 1700, 1521, 1476, 1423, 1215, 1046, 928, 876, 774, 669 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 627.1002 (M^+ + Cs, $C_{21}H_{30}N_6O_6S$ requires 627.1002).

Carbamate 26.[42] *syn*-**18a** (0.014 mmol, 9.1 mg) was subjected to reduction and purification as described for **25** to provide the corresponding alcohol (4.0 mg, 6.6 mg theoretical, 60%) as a white foam. The alcohol (0.009 mmol, 4.0 mg) was subjected to carbamate formation in the same fashion as **25** to give **26** (1.8 mg, 4.5 mg theoretical, 42%): R_f 0.32 (10% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ +18.0 (*c* 0.05, $CHCl_3$); ^1H NMR (CD_3OD, 400 MHz) δ 4.77 (1H, m), 4.66 (1H, d, *J* = 5.6 Hz, C4-H), 4.32 (2H, q, *J* = 7.2 Hz), 4.21 (1H, dd, *J* = 4.9, 10.3 Hz), 3.75 (1H, dd, *J* = 8.2, 10.3 Hz), 3.53 (1H, dd, *J* = 4.1, 13.0 Hz), 3.48 (1H, m), 3.01 (1H, dd, *J* = 8.3, 13.0 Hz), 2.07 (3H, s), 2.05 (3H, s), 1.34 (9H, s), 1.31 (3H, t, *J* = 7.2 Hz); IR ($CHCl_3$) ν_{max} 3010, 2975, 2399 (s, C≡N), 1732, 1699, 1520, 1476, 1423, 1210, 1045, 928 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 627.1021 (M^+ + Cs, $C_{21}H_{30}N_6O_6S$ requires 627.1002).

Carbamate 27.[42] *anti*-**18b** (0.048 mmol, 32 mg) was subjected to reduction and purification as described for **25** to provide the corresponding alcohol as a white foam. The alcohol (0.009 mmol, 4.2 mg) was subjected to carbamate formation in the same fashion as **25** to provide **27** (3.2 mg, 4.1 mg theoretical, 78%): ^1H NMR (CD_3OD, 400 MHz) δ 4.80 (1H, m), 4.72 (1H, d, *J* = 1.2 Hz, C4-H), 4.46 (1H, dd, *J* = 1.8, 12.0 Hz), 4.33 (2H, q, *J* = 7.2 Hz), 4.07 (1H, dd, *J* = 1.5, 12.0 Hz), 3.87 (1H, dd, *J* = 4.5, 14.0 Hz), 3.37 (1H, br s), 3.05 (1H, dd, *J* = 4.8, 14.0 Hz), 2.17 (3H, s), 2.06 (3H, s), 1.38 (9H, s), 1.28 (1H, t, *J* = 7.2 Hz).

Methyl 3(*S*)-(6-Amino-4-(methoxycarbonyl)-5-methylpyrimidin-2-yl)-3-aminopropionate (31). From **20**: Compound **20** (0.011 mmol, 5.2 mg) was treated with 6 N HCl (0.7 mL), and the mixture was stirred at 105 °C for 20 h. The solvent was evaporated in vacuo and the oily solid was treated with 3 N HCl–CH_3OH (1 mL), and the mixture was stirred at 80 °C for 1 h. After evaporation of the solvent, the residue was subjected to chromatography (SiO_2, 1 × 2 cm, 20% CH_3OH–$CHCl_3$) to afford **31** (2.2 mg, 2.9 mg theoretical, 75%): R_f 0.32 (25% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ +3.4 (*c* 0.15, 1 N HCl), $[\alpha]^{25}_{365}$ +36.7 (*c* 0.15, 1 N HCl); ^1H NMR (CD_3OD, 400 MHz) δ 4.66 (1H, dd, *J* = 6.4, 7.2 Hz), 3.96 (3H, s), 3.71 (3H, s), 3.16 (1H, dd, *J* = 6.4, 16.4 Hz), 3.10 (1H, dd, *J* = 7.2, 16.4 Hz), 2.18 (3H, s); IR (neat) ν_{max} 3445, 1715, 1630, 1214, 1143 cm^{-1}; FABHRMS (NBA) *m/e* 269.1255 (M^+ + H, $C_{11}H_{16}N_4O_4$ requires 269.1250).

From bleomycin A_2: A solution of bleomycin A_2 (0.043 mmol, 5.0 mg) was subjected to the same conditions detailed above to provide **31** (0.5 mg, 1.2 mg theoretical, 41%) identical to the material detailed above: $[\alpha]^{25}_{365}$ +26 (*c* 0.15, 1 N HCl).

3(*S*)-(6-Amino-4-carboxy-5-methylpyrimidin-2-yl)-3-aminopropionic Acid (30). The compound **31** (0.007 mmol, 1.8 mg, from **20**) was treated with aqueous 3 N HCl (0.3 mL), and the mixture was stirred at 100 °C for 1 h. The solvent was evaporated in vacuo to afford **30** (1.5 mg, 1.6 mg theoretical, 94%): $[\alpha]^{25}_{365}$ +39.6 (*c* 0.07, 1 N HCl); ^1H NMR (D_2O, 400 MHz) δ 4.94 (1H, m), 3.24 (1H, dd, *J* = 5.8, 16.2 Hz), 3.10 (1H, dd, *J* = 9.0, 16.2 Hz), 2.25 (3H, s).

The sample of **31** derived from bleomycin A_2 similarly provided **30**: $[\alpha]^{25}_{365}$ +30 (*c* 0.2, 1 N HCl).

Methyl 3(*R*)-(6-Amino-4-(methoxycarbonyl)-5-methylpyrimidin-2-yl)-3-aminopropionate (32). Compound **23** (0.012 mmol, 5.7 mg) was subjected to chemical degradation followed by esterification as described for **2D** and bleomycin A_2 to provide **32** (2.1 mg, 3.3 mg theoretical, 64%) identical to **31** except for rotation: $[\alpha]^{25}_D$ –3.7 (*c* 0.14, 1 N HCl), $[\alpha]^{25}_{365}$ –39.3 (*c* 0.14, 1 N HCl).

3(*R*)-(6-Amino-4-carboxy-5-methylpyrimidin-2-yl)-3-aminopropionic Acid (33). Compound **32** (0.0075 mmol, 2.0 mg) was subjected to hydrolysis as described for **31** to provide **33** (1.7 mg, 1.8 mg theoretical, 94%) identical to **30** except for rotation: $[\alpha]^{25}_{365}$ –40.3 (*c* 0.08, 1 N HCl).

Subjection of **39** to the same hydrolysis conditions also provided **33** identical to that detailed above but of higher enantiomeric purity: $[\alpha]^{25}_{365}$ –59 (*c* 0.11, 1 N HCl).

Ethyl 2-[1(*R*)-Hydroxy-2(*S*)-(methylthio)-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (35). A solution of (4*S*,5*R*)-3-((methylthio)acetyl)-4-methyl-5-phenyl-2-oxazolidinone (**34**, 0.15 mmol, 38 mg) in dry CH_2Cl_2 (0.7 mL) under N_2 at –78 °C was treated with Bu_2BOTf (0.15 mmol, 41 mg, 33 μL, 1.0 equiv) followed by iPr_2NEt (0.165 mmol, 21 mg, 29 μL, 1.1 equiv). The mixture was stirred at 0 °C for 1 h and recooled to –78 °C. A solution of **15** (0.038 mmol, 7.8 mg, 0.25 equiv) in dry CH_2Cl_2 (0.2 mL) was added, and the reaction mixture was allowed to warm to 0 °C, where it was stirred for 3 h. The reaction mixture was poured into a two-phase solution of CH_2Cl_2 (2 mL) and saturated aqueous $NaHCO_3$ (2 mL) with vigorous stirring. The organic layer was washed with saturated aqueous NaCl (2 × 1 mL), dried over (Na_2SO_4), and concentrated in vacuo. Chromatography (SiO_2, 1 × 4 cm, 2% CH_3OH–CH_2Cl_2) gave **35** as a white foam (10 mg, 18 mg theoretical, 59%): R_f 0.32 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ –8.0 (*c* 0.52, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.40–7.20 (5H, m), 5.75 (1H, d, *J* = 7.2 Hz), 5.30 (2H, br s), 5.26 (1H, d, *J* = 8.8 Hz), 5.13 (1H, dd, *J* = 4.0, 8.8 Hz), 4.83 (2H, q, *J* = 6.8 Hz), 4.39 (2H, m), 2.23 (3H, s), 2.20 (3H, s), 1.40 (3H, t, *J* = 6.0 Hz), 0.92 (3H, d, *J* = 6.8 Hz); IR ($CHCl_3$) ν_{max} 3419, 3018, 1778, 1729, 1690, 1612, 1570, 1521, 1443, 1368, 1213, 1671, 928, 756 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 607.0632 (M^+ + Cs, $C_{22}H_{26}N_4O_6S$ requires 607.0627).

Ethyl 2-[1(*S*)-Hydroxy-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (36). A solution of **35** (0.044 mmol, 20 mg) in C_6H_6 (0.7 mL) was treated with Bu_3SnH (0.13 mmol, 38 mg, 3.0 equiv) and AIBN (0.013 mmol, 3 mg, 0.3 equiv) and warmed at 80 °C for 1 h under N_2. The mixture was allowed to cool to 23 °C, and the solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 4 cm, 5% CH_3OH–CH_2Cl_2) afforded **36** as a white foam (15.6 mg, 17 mg theoretical, 93%): R_f 0.32 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ –37.2 (*c* 0.42, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.40–7.20 (5H, m), 5.91 (2H, br s), 5.70 (1H, d, *J* = 7.2 Hz), 5.15 (1H, dd, *J* = 3.3, 8.0 Hz), 4.80 (1H, dq, *J* = 6.8, 7.2 Hz), 4.41 (2H, q, *J* = 6.8 Hz), 3.61 (1H, dd, *J* = 3.6, 16.4 Hz), 3.47 (1H, dd, *J* = 8.0, 16.4 Hz), 2.19 (3H, s), 1.39 (3H, t, *J* = 6.8 Hz), 0.90 (1H, d, *J* = 6.8 Hz); IR ($CHCl_3$) ν_{max} 3420, 3018, 1780, 1726, 1612, 1521, 1424, 1214, 1043, 928 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 561.0747 (M^+ + Cs, $C_{21}H_{24}N_4O_6$ requires 561.0750).

Ethyl 2-[1(*R*)-Azido-2-[((4*S*,5*R*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (37). Diethyl azodicarboxylate (0.043 mmol, 7.5 mg, 7.0 μL, 1.2 equiv) and a 0.8 M benzene solution of hydrazoic acid[43] (0.043 mmol, 54 μL, 1.2 equiv) were added sequentially to a solution of **36** (0.036 mmol, 15 mg) and Ph_3P (0.043 mmol, 12 mg, 1.2 equiv) in dry THF (0.5 mL) at –78 °C. The mixture was stirred for 16 h at 23 °C. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 6 cm, 2% CH_3OH–CH_2Cl_2) afforded **37** as a foam (8.2 mg, 15.7 mg theoretical, 52%): R_f 0.42 (5% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +35.6 (*c* 0.53, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.30–7.10 (5H, m), 5.65 (1H, d, *J* = 7.2 Hz), 5.14 (2H, br s), 5.00 (1H, dd, *J* = 4.8, 8.4 Hz), 4.75 (1H, dq, *J* = 6.8, 7.2 Hz), 4.37 (2H, q, *J* = 7.2 Hz), 3.53 (1H, d, *J* = 4.8 Hz), 3.50 (1H, d, *J* = 8.4 Hz), 2.14 (3H, s), 1.35 (3H, t, *J* = 7.2 Hz), 0.83 (3H, d, *J* = 6.8 Hz); IR ($CHCl_3$) ν_{max} 3018, 2107, 1723, 1709, 1611, 1519, 1473, 1422, 1388,

(42) Reduction with excess $NaBH_4$ (EtOH) at 25 °C led to C4 ethyl ester reduction. Initial attempts to form the cyclic carbamates upon reaction with diethyl carbonate (K_2CO_3) and methyl chloroformate (C_6H_6, pyridine, 25 °C, 30 min, 79%) provided corresponding uncyclized methyl carbamate which failed to close to **25**–**27** upon further treatment with base (DMAP, K_2CO_3, DBU, NaH; C_6H_6, DMF, THF; 25–80 °C). Similarly, treatment with carbonyldiimidazole (C_6H_6, 80 °C) provided the *N*-carbonylimidazole, which failed to close cleanly to the desired cyclic carbamate.

(43) Wolff, H. *Org. React.* **1946**, *3*, 327.

1218, 1045, 928, 757 cm^{-1}; FABHRMS (NBA) *m/e* 454.1830 (M^+ + H, $C_{21}H_{23}N_7O_5$ requires 454.1839).

Methyl 3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*R*)-azidopropionate (38). A solution of **37** (0.018 mmol, 8.0 mg) in CH_3OH (0.3 mL) under N_2 at 0 °C was treated with $NaOCH_3$ (0.018 mmol, 1.0 mg, 1.0 equiv), and the mixture was stirred at 0 °C for 20 min. The reaction mixture was poured into a two-layer solution of CH_2Cl_2 (2 mL) and saturated aqueous NaCl (1 mL) with vigorous stirring. The organic layer was washed with saturated aqueous NaCl (1 mL), dried (Na_2SO_4), and concentrated in vacuo. Chromatography (SiO_2, 1 × 3 cm, 2% CH_3OH–CH_2Cl_2) gave **38** as a film (3.2 mg, 5.3 mg theoretical, 59%): R_f 0.35 (20% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +61.6 (*c* 0.19, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 5.13 (2H, br s), 4.90 (1H, dd, *J* = 5.2, 8.8 Hz), 4.42 (2H, q, *J* = 6.8 Hz), 3.73 (3H, s), 3.04 (1H, dd, *J* = 5.2, 17.2 Hz), 2.87 (1H, dd, *J* = 8.8, 17.2 Hz), 2.21 (3H, s), 1.41 (3H, t, *J* = 6.8 Hz); IR ($CHCl_3$) ν_{max} 3017, 2392, 1722, 1508, 1364, 1242, 1208, 1046, 924, 843, 745 cm^{-1}; FABHRMS (NBA) *m/e* 309.1310 (M^+ + H, $C_{12}H_{16}N_6O_4$ requires 309.1311).

Methyl 3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*R*)-aminopropionate (39). A solution of **38** (0.008 mmol, 25 mg) in THF (0.3 mL) was treated with Ph_3P (0.01 mmol, 4.5 mg, 2.0 equiv) in the presence of H_2O (5 μL), and HOAc (5 μL) and the mixture was stirred under N_2 at 25 °C for 15 h. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 2 cm, 20% CH_3OH–CH_2Cl_2) afforded **39** as a white film (1.9 mg, 2.3 mg theoretical, 82%): R_f 0.12 (5% CH_3OH–$CHCl_3$); $[\alpha]^{25}_D$ −5.6 (*c* 0.16, 1 N HCl), $[\alpha]^{25}_{365}$ −53.8 (*c* 0.16, 1 N HCl); ^1H NMR (CD_3OD, 400 MHz) δ 4.77 (1H, br s), 4.27 (2H, q, *J* = 7.2 Hz), 3.49 (3H, s), 3.00 (1H, dd, *J* = 5.6, 17.4 Hz), 2.87 (1H, dd, *J* = 7.2, 17.4 Hz), 2.19 (3H, s), 1.19 (3H, t, *J* = 7.2 Hz); IR ($CHCl_3$) ν_{max} 3329, 2936, 1721, 1653, 1450, 1414, 1213, 1023, 924, 739 cm^{-1}; FABHRMS (NBA) *m/e* 283.1400 (M^+ + H, $C_{12}H_{18}N_4O_4$ requires 283.1406).

3(*R*)-(6-Amino-4-carboxy-5-methylpyrimidin-2-yl)-3-aminopropionic Acid (33). The compound **39** (0.0087 mmol, 2.4 mg) was treated with 3 N aqueous HCl (0.3 mL), and the mixture was stirred at 100 °C for 1 h. The solvent was evaporated in vacuo to afford **33** (2.0 mg, 2.2 mg theoretical, 91%): $[\alpha]^{25}_{365}$ −59.3 (*c* 0.11, 1 N HCl); ^1H NMR (D_2O, 400 MHz) δ 4.94 (1H, m), 3.24 (1H, dd, *J* = 5.2, 17.1 Hz), 3.13 (1H, dd, *J* = 7.2, 17.1 Hz), 2.25 (3H, s); IR (neat) ν_{max} 3438, 2921, 1708, 1646, 1527, 1441, 1216, 1051, 865 cm^{-1}; FABHRMS (NBA) *m/e* 241.0930 (M^+ + H, $C_9H_{12}N_4O_4$ requires 241.0937).

Ethyl 2-[1(*S*)-Hydroxy-2(*R*)-(methylthio)-2-[((4*R*,5*S*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (41). A solution of (4*R*,5*S*)-3-((methylthio)acetyl)-4-methyl-5-phenyl-2-oxazolidinone (**40**, 2.30 mmol, 576 mg) in dry CH_2Cl_2 (2 mL) under N_2 at −78 °C was treated with Bu_2BOTf (2.30 mmol, 630 mg, 1.0 equiv) followed by iPr_2NEt (2.52 mmol, 326 mg, 0.44 mL, 1.1 equiv). The mixture was stirred at 0 °C for 1 h, and the reaction mixture was recooled to −78 °C. A solution of **15** (0.57 mmol, 120 mg, 0.25 equiv) in dry CH_2Cl_2 (2 mL) was added, and the reaction mixture was allowed to warm to 0 °C where it was stirred for 3 h. The reaction mixture was poured into a two-phase solution of CH_2Cl_2 (20 mL) and saturated aqueous $NaHCO_3$ (10 mL) with vigorous stirring. The organic layer was washed with saturated aqueous NaCl (2 × 5 mL), dried (Na_2SO_4), and concentrated in vacuo. Chromatography (SiO_2, 1 × 8 cm, 2% CH_3OH–CH_2Cl_2) gave **41** as a thin film (180 mg, 274 mg theoretical, 67%): R_f 0.32 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +7.8 (*c* 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.40–7.20 (5H, m), 5.75 (1H, d, *J* = 7.2 Hz), 5.30 (2H, br s), 5.26 (1H, d, *J* = 8.8 Hz), 5.13 (1H, dd, *J* = 4.0, 8.8 Hz), 4.83 (2H, q, *J* = 6.8 Hz), 4.77 (1H, dq, *J* = 6.8, 7.2 Hz), 4.39 (2H, m), 2.23 (3H, s), 2.20 (3H, s), 1.40 (3H, t, *J* = 6.8 Hz), 0.92 (3H, d, *J* = 6.8 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.5, 165.9, 165.2, 163.5, 153.2, 152.6, 133.7, 128.6, 125.7, 125.6, 111.1, 78.7, 73.8, 71.2, 62.0, 54.6, 48.9, 14.5, 14.1, 13.2, 11.9; IR ($CHCl_3$) ν_{max} 3419, 3018, 1778, 1729, 1690, 1612, 1570, 1521, 1443, 1368, 1213, 1071, 928, 756 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 607.0632 (M^+ + Cs, $C_{22}H_{26}N_4O_6S$ requires 607.0627).

Ethyl 2-[1(*R*)-Hydroxy-2-[((4*R*,5*S*)-4-methyl-5-phenyl-2-oxazolidinyl)carbonyl]ethyl]-6-amino-5-methylpyrimidine-4-carboxylate (42). A solution of **41** (0.043 mmol, 20 mg) in C_6H_6 (0.7 mL) was treated with Bu_3SnH (0.13 mmol, 38 mg, 3.0 equiv) and warmed at 80 °C for 1 h under N_2. The mixture was allowed to cool to 23 °C, and the solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 3 cm, 5% CH_3OH–CH_2Cl_2) afforded **42** as a white film (16 mg, 17 mg theoretical, 93%): R_f 0.32 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +37.5 (*c* 0.13, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.40–7.20 (5H, m), 5.91 (2H, br s), 5.70 (1H, d, *J* = 7.2 Hz), 5.15 (1H, dd, *J* = 3.3, 8.0 Hz), 4.80 (1H, dq, *J* = 6.8, 7.2 Hz), 4.41 (2H, q, *J* = 6.8 Hz), 3.61 (1H, dd, *J* = 3.6, 16.4 Hz), 3.47 (1H, dd, *J* = 8.0, 16.4 Hz), 2.19 (3H, s), 1.39 (3H, t, *J* = 6.8 Hz), 0.90 (1H, d, *J* = 6.8 Hz); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 170.6, 166.2, 165.8, 162.9, 153.1 (2C), 133.2, 128.6, 128.5, 125.6, 110.6, 79.0, 69.0, 61.7, 54.6, 42.6, 14.3, 14.0, 11.5; IR ($CHCl_3$) ν_{max} 3420, 3018, 1780, 1726, 1612, 1521, 1424, 1214, 1043, 928 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 561.0747 (M^+ + Cs, $C_{21}H_{24}N_4O_6$ requires 561.0750).

3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*R*)-hydroxypropionamide (43). The agent **42** (0.14 mmol, 57 mg) was treated with an ethanolic solution of NH_3 (16%, 8 mL), and the solution was stirred for 1.5 h at 0 °C. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 5 cm, 10% CH_3OH–CH_2Cl_2) afforded **43** as a white film (27 mg, 35 mg theoretical, 77%): R_f 0.12 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ +35 (*c* 0.10, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 4.82 (1H, br s), 4.30 (2H, q, *J* = 7.2 Hz), 2.67 (1H, dd, *J* = 3.6, 14.8 Hz), 2.50 (1H, dd, *J* = 8.8, 14.8 Hz), 2.04 (3H, s), 1.28 (3H, t, *J* = 7.2 Hz); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 176.2, 168.2, 167.7, 165.7, 154.3, 71.9, 63.1, 43.5, 14.5, 12.3; IR ($CHCl_3$) ν_{max} 3423, 3018, 1728, 1676, 1522, 1424, 1213, 1046, 928 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 401.0224 (M^+ + Cs, $C_{11}H_{16}N_4O_4$ requires 401.0226).

3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*S*)-azidopropionamide (44). Diethyl azodicarboxylate (0.19 mmol, 33 mg, 1.2 equiv) and a 0.8 M benzene solution of hydrazoic acid[43] (0.48 mmol, 0.60 mL, 3 equiv) were added sequentially to a solution of **43** (0.16 mmol, 41 mg) and Ph_3P (0.19 mmol, 51 mg, 1.2 equiv) in dry THF (1.2 mL) at −78 °C. The mixture was stirred for 36 h at 0 °C. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 1 × 5 cm, 5% CH_3OH–CH_2Cl_2) afforded **44** as a white film (38 mg, 45 mg theoretical, 84%): R_f 0.45 (10% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ −71.7 (*c* 0.12, $CHCl_3$); ^1H NMR (CD_3OD, 400 MHz) δ 4.65 (1H, dd, *J* = 5.6, 8.8 Hz), 4.31 (2H, q, *J* = 7.2 Hz), 2.80 (1H, dd, *J* = 5.2, 14.8 Hz), 2.66 (1H, dd, *J* = 9.6, 14.8 Hz), 2.05 (3H, s), 1.29 (3H, t, *J* = 7.2 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 175.0, 167.5, 165.8, 165.1, 154.8, 112.0, 63.6, 63.0, 39.7, 14.4, 12.1; IR ($CHCl_3$) ν_{max} 3421, 3018, 2107, 1724, 1522, 1424, 1218, 928 cm^{-1}; FABHRMS (NBA) *m/e* 294.1313 (M^+, $C_{11}H_{15}N_7O_3$ requires 294.1315).

3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*S*)-aminopropionamide (45). A solution of **44** (0.09 mmol, 25 mg) in CH_3OH (4 mL) was stirred with 10% Pd-C (6 mg) for 1 h under a H_2 atmosphere. The catalyst was removed by filtration through Celite, and the filtrate was concentrated in vacuo. Flash chromatography (SiO_2, 1 × 3 cm, 20% CH_3OH–CH_2Cl_2) afforded **45** as a film (20 mg, 23 mg theoretical, 88%): R_f 0.10 (20% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ −20.0 (*c* 0.12, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 4.31 (2H, q, *J* = 6.8 Hz), 4.14 (1H, br s), 2.74 (1H, dd, *J* = 4.0, 15.2 Hz), 2.49 (1H, dd, *J* = 8.4, 15.2 Hz), 2.04 (3H, s), 1.28 (3H, t, *J* = 6.8 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.5, 169.1, 167.8, 165.7, 154.6, 111.0, 63.0, 55.0, 43.1, 14.4, 12.0; IR (neat) ν_{max} 3394, 2984, 1718, 1656, 1579, 1446, 1405, 1236, 1077, 1015 cm^{-1}; FABHRMS (NBA–NaI) *m/e* 290.1220 (M^+ + Na, $C_{11}H_{17}N_5O_3$ requires 290.1229).

3-(6-Amino-4-(ethoxycarbonyl)-5-methylpyrimidin-2-yl)-3(*S*)-[(2-(((*tert*-butyloxy)carbonyl)amino)ethyl)amino]propionamide (46). A solution of **45** (0.02 mmol, 5.0 mg) and *N*-((*tert*-butyloxy)carbonyl)-2-bromoethylamine (0.04 mmol, 9.0 mg, 2.0 equiv) in DMF (45 μL) was treated with K_2CO_3 (0.025 mmol, 3.5 mg, 1.2 equiv), and the mixture was stirred under Ar at 25 °C for 2 days. The solvent was evaporated in vacuo. Flash chromatography (SiO_2, 10% CH_3OH–CH_2Cl_2) afforded **45** as a film (4.2 mg, 8.2 mg theoretical, 51%): R_f 0.38 (20% CH_3OH–CH_2Cl_2); $[\alpha]^{25}_D$ −22.0 (*c* 0.075, CH_3OH); ^1H NMR (CD_3OD, 400 MHz) δ 4.29 (2H, q, *J* = 6.8 Hz), 3.86 (1H, m), 3.02 (2H, m), 2.45 (2H, m), 2.02 (3H, s), 1.28 (9H, s), 1.26 (3H, t, *J* = 6.8 Hz); ^{13}C NMR (CD_3OD, 100 MHz) δ 176.4, 168.1, 167.8, 165.9, 158.5, 154.5, 80.2, 63.1, 61.6, 48.0, 41.3, 41.1, 28.9, 14.5, 12.1; IR (neat) ν_{max} 3420, 1718, 1638, 1581, 1439, 1259, 865 cm^{-1}; FABHRMS (NBA–CsI) *m/e* 543.1340 (M^+ + Cs, $C_{18}H_{30}N_6O_5$ requires 543.1332).

3-(6-Amino-4-carboxy-5-methylpyrimidin-2-yl)-3(*S*)-[(2-(((*tert*-butyloxy)carbonyl)amino)ethyl)amino]propionamide (47). A solution of **46** (0.01 mmol, 4.2 mg) in THF–CH_3OH–H_2O (3:1:1, 0.3 mL) at 0 °C was treated with aqueous 1 N LiOH (0.015 mmol, 15 μL, 1.5 equiv), and the mixture was stirred for 1.5 h. After evaporation of most of the THF–CH_3OH, the aqueous phase was acidified to pH 4–5 with the addition of aqueous 1.2 N HCl and the solvent was evaporated in vacuo. The residue was charged onto a column of Dowex (1 × 2 cm; acetate form, 50–100 mesh). The column was washed with H_2O, and subsequent elution with 6% HOAc–H_2O afforded **47** as a thin film (3.3 mg, 3.9 mg theoretical,

85%): R_f 0.62 (4:1:1 *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ –8.0 (*c* 0.20, H_2O); ^1H NMR (CD_3OD, 400 MHz) δ 4.20 (1H, br s), 3.18 (2H, m), 2.87 (2H, m), 2.78 (1H, dd, J = 4.4, 16.0 Hz), 2.67 (1H, dd, J = 8.0, 16.0 Hz), 2.02 (3H, s), 1.31 (9H, s); IR (neat) ν_{max} 3333, 2964, 1676, 1574, 1420, 1169 cm^{-1}; FABHRMS (NBA) *m/e* 383.2051 (M^+ + H, $C_{16}H_{26}N_6O_5$ requires 383.2043).

3-(6-Amino-4-carboxy-5-methylpyrimidin-2-yl)-3(*S*)-[(2-aminoethyl)-amino]propionamide ((–)-descarboxamidopyrimidoblamic acid, 4). The compound **47** (0.003 mmol, 1.2 mg) was treated with 3 N HCl–EtOAc (0.3 mL), and the mixture was stirred at 25 °C for 1 h. The solvent was evaporated in vacuo to give pure **4** hydrochloride (0.9 mg, 1.0 mg theoretical, 90%) as a clear hygroscopic solid: R_f 0.38 (4:1:1 *i*PrOH–H_2O–HOAc); $[\alpha]^{25}_D$ –17.8 (*c* 0.045, H_2O); ^1H NMR (D_2O, 400 MHz) δ 4.66 (1H, dd, J = 5.6, 7.2 Hz), 3.36 (4H, m), 3.10 (1H, dd, J = 5.6, 16.8 Hz), 3.02 (1H, dd, J = 7.2, 16.8 Hz), 2.18 (3H, s); IR (neat) ν_{max} 3491, 3199, 1696, 1627, 1542, 1362, 1203 cm^{-1}; FABHRMS (NBA) *m/e* 283.1520 (M^+, $C_{11}H_{18}N_6O_3$ requires 283.1519).

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