Synthesis and Biological Evaluation of Analogues of the Antibiotic Pantocin B

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Abstract: Strains of the bacteria *Erwinia herbicola* produce antibiotics that effectively control *E. amylovora*, the bacterial pathogen responsible for the plant disease fire blight. Pantocin B was the first of these antibiotics to be characterized, and a flexible synthesis of various analogues is reported. Embedded in the "pseudo-tripeptide" backbone of pantocin B are a methylenediamine and a methyl sulfone, both unusual structural features in natural products. The peptidic nature of pantocin B facilitated a series of structure−activity relationship studies that probed the roles of these functional groups in determining the biological activity of pantocin B. A clear demarcation of the roles between the N- and C-terminal portions of the antibiotic was determined as a result of the structure−activity relationship studies. The N-terminal L-alanyl group is needed for cellular import but not for interaction with the intracellular target, the arginine biosynthetic enzyme *N*-acetylornithine aminotransferase. The methylenediamine and methyl sulfone portions were found to be essential for antibiotic activity, presumably due to extensive interactions with *N*-acetylornithine aminotransferase.

Introduction

Fire blight is a devastating disease of pome fruit plants, most notably apple and pear trees, that results from an infection by the pathogenic bacterium *Erwinia amylovora*. A closely related bacterium, *E. herbicola* (syn. *Pantoea agglomerans*) colonizes the same plant surfaces as *E. amylovora* but is not pathogenic to the plant.[1] In fact, *E. herbicola* has been found to produce an array of antibiotics that suppress growth of *E. amylovora* both in the laboratory and the field.[2] The antibiotics produced by *E. herbicola* have different patterns of biological activity and, presumably, chemical structures. One convenient way to distinguish among the antibiotics is the suppression of antibiotic activity by amino acids.[3−5] In the best-studied case, *E. herbicola* strain 318, which produces pantocin A and B, pantocin B's antibiotic activity is suppressed by adding arginine to the growth medium while pantocin A's activity is suppressed by adding histidine.[6] The multiplicity of antibiotics produced by *E. herbicola* has hindered bioassay-guided isolation of individual antibiotics from wild-type strains, and a recent publication described an alternative method, the use of a genomic library and heterologous expression, to facilitate the isolation of natural products arising from such complex biological systems.[6] Construction of a cosmid library of *E. herbicola* 318 and heterologous expression in *E. coli* led to the isolation and characterization of pantocin B, and in this paper we describe our initial efforts to explore the chemistry and biology of pantocin-like antibiotics through synthesis. Pantocin B (**1**), shown in Figure 1, exists as pair of diastereomers due to the ease of epimerization of the sulfone-bearing methine.[6,7] Compound **2** was isolated along with pantocin B and has no measurable antibiotic activity against *E. amylovora* test strains. Since the activity of pantocin B is suppressed by arginine, its target was sought along the arginine biosynthetic pathway. Using a series of specific mutants, pantocin B was shown to be a competitive, with respect to *N*-acetylornithine (**4**), inhibitor of *N*-acetylornithine aminotransferase (Acorn), an enzyme involved in arginine biosynthesis (Figure 2).[6]

Both pantocin B and the inactive isolate **2** contain unusual structural elements, most notably the methylenediamine moiety, which has been reported in the structure of only one other antibiotic.[8] The succinic acid fragment of pantocin B is also not well precedented in natural products. To begin exploring pantocin B's biological activity and its structural dependence a total synthesis of **1** was recently developed (Scheme 1).[7] The synthetic route to pantocin B was designed to allow for a straightforward adaptation of the synthesis to structural analogues. Utilizing such analogues in structure−activity relationship (SAR) studies provides a route to explore the antibiotic's mode of action and gain insight into the roles of the unusual chemical moieties embedded in the structure.

Results and Discussion

Our analysis of pantocin B focused on its two internal amide bonds (Figure 3). This "tripeptide" analysis seemed to provide

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(1) Beer, S. V.; van der Zwet, T. *Fire Blight: Its Nature, Prevention, and Control: A Practical Guide to Integrated Disease Management*; U.S. Department of Agriculture, U.S. Government Printing Office: Washington, DC, 1995.

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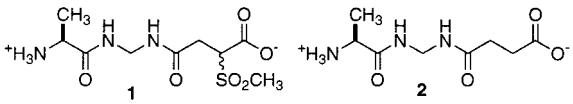

Figure 1. Pantocin B and related natural product.

Figure 2. Reaction catalyzed by *N*-acetylornithine aminotransferase (Acorn).

the easiest route to independently vary the structural features of pantocin B to provide analogues.

The N-Terminal Analogues. The synthesis of the N-terminal analogues proved to be straightforward. By utilizing the general synthetic route developed for pantocin B (Scheme 1) with a number of benzyloxycarbonyl (*Z*)-protected amino acids, a series of structural analogues of pantocin B were synthesized (Figure 4). The synthesis of pantocin B is convergent, coupling the alanyl intermediate, **8a**, with the succinyl intermediate, **13a**, to construct the backbone of the antibiotic. Utilizing Loudon's methodology,[9] the desired methylenediamine is synthesized via an "acidic Hofmann" rearrangement from the glycinamide **7a**. The succinyl derivative is synthesized from L-malic acid in four steps, utilizing a triflate in the key substitution step to introduce the desired thioether moeity. Following the coupling of **8a** and **13a**, the oxidation of the methyl sulfide in **14a** gave rise to the desired sulfone moiety present in pantocin B. After a simple hydrogenation to remove the protecting groups, the desired zwitterion, **1**, was synthesized. The substitution of the *Z*-L-alanine with a series of protected amino acids in this convergent synthesis readily gave rise to the desired N-terminal analogues. The overall yields of final derivatives, **16**−**22**, from the *Z*-amino acids ranged from 5 to 17%.

All of the analogues were assayed against *E. amylovora* utilizing a disk diffusion assay. Antibiotic activity was determined by zones of inhibited growth measured at compound loading of 125 ng, 63 ng, and 31 ng. All of the L-amino acid analogues had an arginine-suppressible antibiotic activity essentially identical to that of pantocin B (Table 1); only the L-prolyl and L-phenylalanyl analogues exhibited antibiotic activity at reduced levels compared to pantocin B. None of the D-amino acid analogues, including the D-alanyl derivative (**21**), had any detectable antibiotic activity. This result—activity for any L-amino acid, no activity for any D-amino acid—suggests that access to a cell via a peptide transporter is essential for antibiotic activity.

Peptide transporters, permeases which regulate the uptake of peptides into cells, in general, exhibit high stereoselectivity but low substrate specificity.[10] The broad substrate specificity inherent in peptide transporters has presumably evolved to handle the structural diversity intrinsic in the side chains of protein-derived oligopeptides.[11] While in some cases the only feature needed for successful transport is a zwitterionic amino acid functionality separated by an ideal distance,[12] in most transporters the N-terminal α-amino group plays the crucial role in substrate recognition.[11] The effective transport of oligopeptides requires a positively charged primary or secondary α-amino group. The C-terminal end is less critical, and a greater degree of structural variation, including nonpeptidic elements and D-amino acids, can be tolerated.[11]

An investigation to probe the role of peptide transport in determining the bioactivity of pantocin B and analogues was undertaken through an antibiotic activity suppression study. Since pantocin B is similar in size to a tripeptide, it is most likely transported into *E. amylovora* via the oligopeptide permease (Opp).[11] At sufficiently high concentrations of an appropriate tripeptide, Opp should be saturated with added tripeptide and unavailable to transport pantocin B and active analogues. The antibiotic activity of pantocin B and L-amino acid analogues were determined in the presence of the tripeptides L-ala·gly·gly and D-ala·gly·gly added in varying amounts (10^{-2}−10^{-6} M) to the test media (Table 2). As expected L-ala·gly·gly suppressed the antibiotic activities of both pantocin B and the active analogues at relatively high concentrations. The inability of D-ala·gly·gly to suppress antibiotic activity suggests that peptide transport is an essential prerequisite for antibiotic activity.

The Mid-Section Analogues. To probe the effect of substitution on the central methylene of pantocin B, two analogues were synthesized employing L- or D-alanamide hydrochloride rather than glycinamide hydrochloride, **6**, in the general synthesis developed for pantocin B (Scheme 1). The two analogues (**23**, **24**) were synthesized in 6 and 25% overall yield from the L-alanamide hydrochloride and D-alanamide hydrochloride, respectively (Figure 5).

To investigate the role of the chain length, a derivative with an additional methylene was synthesized. The synthesis of **25** differed significantly from that of pantocin B (Scheme 2). The *Z*-L-alanine was coupled to **26** utilizing 1,1′-carbonyldiimidazole as a coupling agent in 61% yield. After selective deprotection of the Boc group, the desired amine hydrochloride **27** could be coupled to **13a** utilizing the methodologies developed for the synthesis of pantocin B. The desired analogue **25** was synthesized from **29** in 34% yield.

All three mid-section analogues proved to be inactive against *E. amylovora* (Table 3). The inactivity of all three analogues indicates a central unsubstituted methylenediamine is essential for pantocin B's activity. The demonstrated tolerance of most peptide transporters[11,12] for modifications away from the N-terminus suggests the substituted central methylene does not interfere with transport into *E. amylovora*, but lack of recognition of these analogues by the Opp transporter cannot be ruled out.

The C-Terminal Analogues. The inactive natural product **2** isolated along with pantocin B indicated that changes in the C-terminal portion would affect antibiotic activity. The first set of C-terminal analogues had altered oxidation states for the sulfur atom. These analogues were synthesized with only minimal modifications to the synthesis of pantocin B (Scheme 3). The utilization of methanolic formic acid solution in the transfer hydrogenation successfully synthesized the desired thioether analogues (**30**, **31**) in moderate to good yields despite the presence of an unoxygenated sulfur.[13] The desired sulfoxide was readily synthesized employing 1 equiv of mCPBA in the oxidation step. The resultant sulfoxide was directly deprotected via a transfer hydrogenation utilizing methanolic formic acid to yield **32** in 56% yield.

(9) Loudon, G. M.; Radhakrishna, A. S.; Almond, M. R.; Blodgett, J. K.; Boutin, R. H. *J. Org. Chem.* **1984**, *49*, 4272−4276.

(10) Steiner, H.-Y.; Naider, F.; Becker, J. M. *Mol. Microbiol.* **1995**, *16*, 825−834.

(11) Payne, J. W. In *Peptide-Based Drug Design*; Taylor, M. D., Amidon, G. L., Eds.; American Chemical Society: Washington, DC, 1995; pp 341−367.

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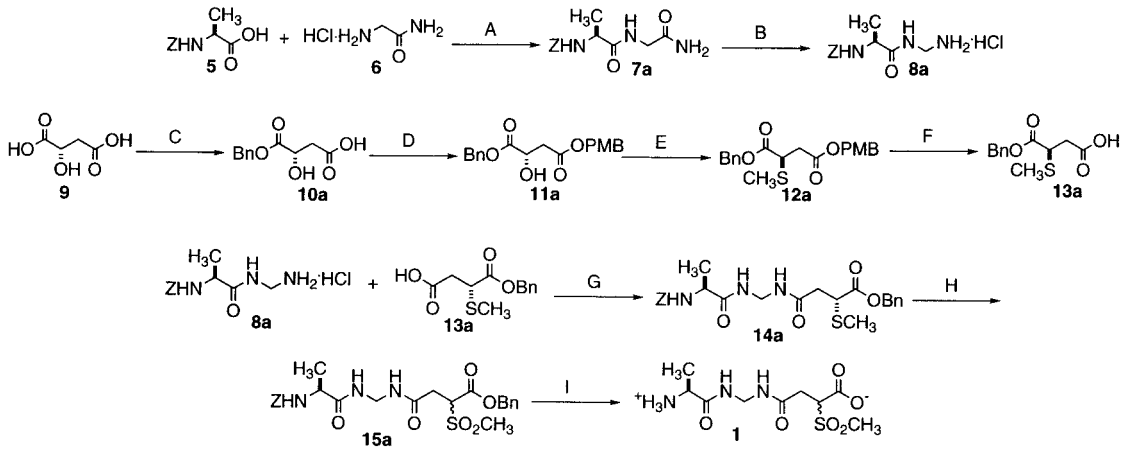
(13) ElAmin, B.; Anantharamaiah, G. M.; Royer, G. P.; Means, G. E. *J. Org. Chem.* **1979**, *44*, 3442−3444.

Scheme 1[a]



[a] (A) CDI, NEt_3, THF. (B) i) [*I*,*I*-bis(trifluoroacetoxy)iodo]benzene (PIFA), aq MeCN, ii) 1 N HCl. (C) i) TFAA, 0 °C, ii) BnOH. (D) 4-methoxybenzyl chloride, K_2CO_3, DMF. (E) i) Tf_2O, 2,6-lutidine, CH_2Cl_2, −78 °C, ii) CH_3SNa, 15-crown-5, DMF, −78 °C to rt. (F) 10% TFA in CH_2Cl_2, 0 °C. (G) EDAC, HOBT, *N*-ethylmorpholine, CH_2Cl_2. (H) mCPBA, CH_2Cl_2, 0 °C. (I) Pd-black, 5% HCOOH in MeOH.

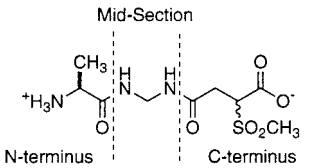


Figure 3. Structural sections of pantocin B.

The second set of C-terminal analogues were designed in order to investigate whether the sulfone moiety functioned as a leaving group to generate a unsaturated molecule that could interact with the enzyme target via a conjugate addition (Scheme 4).

The synthesis of acrylic acid containing analogues required a modified synthetic approach. In particular, the choice of protecting groups needed to allow for selective cleavage without disrupting the unsaturation present in the analogue (Scheme 5). The Fmoc group was chosen due its ability to be cleaved under conditions that would both preserve the double bonds and be stable under the acidic conditions needed to effect the rearrangement to the amine hydrochloride **35**.[9] After some experimentation, diisopropylethylamine was chosen as the tertiary amine base present in the coupling reactions as only trace cleavage of the Fmoc group was observed during the course of the coupling. The desired glycinamide derivative could be synthesized in 50% yield from Fmoc-L-alanine. After rearrangement the resultant amine hydrochloride **35** could be coupled with maleic anhydride, which after deprotection, resulted in the desired *Z*-acrylic acid derivative (**38**). Similarly, **35** could be coupled with **39** in 70% yield. After two in situ deprotection steps the resultant *E*-acrylic acid derivative (**41**) was synthesized in 46% yield.

The C-terminal analogues were assayed against *E. amylovora*, and interestingly, only the sulfoxide derivative had antibiotic activity (Table 4). The activity of the sulfoxide demonstrated the importance of the oxygenated center, while the inactivity of both acrylic acid derivatives indicated that the sulfone moiety was probably not serving as a leaving group to create an unsaturated biologically active molecule. Both the sulfone and sulfoxide groups are commonly employed in medicinal chemistry as bioisosteres for the carbonyl group.[14] Since pantocin B was demonstrated to be a competitive inhibitor of *N*-acetylornithine (**4**),[6] the sulfone and the sulfoxide moieties may serve as bioisosteres for the *N*-acetyl group present in **4** (Figure 6). *N*-acetylornithine aminotransferase, a class II PLP-dependent aminotransferase, has been demonstrated to have a broad substrate specificity, especially with regard to the amino donor. In addition a variety of α-*N*-acyl substituents can be accommodated by the enzyme, including substituents as large as the carbobenzyloxy group.[15] Acorn's broad substrate specificity

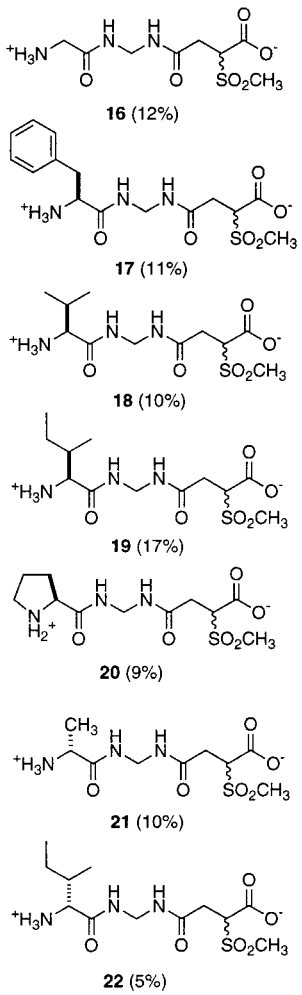


Figure 4. N-terminal analogues.

(14) Thornber, C. W. *Chem. Soc. Rev.* **1979**, *8*, 563−580.

(15) Ledwidge, R.; Blanchard, J. S. *Biochemistry* **1999**, *38*, 3019−3024.

Table 1. Antibiotic Activity of N-Terminal Analogues against *E. amylovora*[a]

analogue	125 ng[b]	63 ng[b]	31 ng
pantocin B	19.3	16.5	14.0
16	19	17	14
17	<16	<14	0
18	18	16	12
19	19	16	14
20	<13	<12	0
21	0	0	0
22	0	0	0

[a] Kill zones are given in mm. [b] < denotes a hazy inhibition zone.

Table 2. Tripeptide Antibiotic Suppression Studies[a]

analogue (125 ng)	L-ala-gly-gly (10^{-2} M)	L-ala-gly-gly (10^{-4} M)	L-ala-gly-gly (10^{-6} M)	D-ala-gly-gly (10^{-2} M)
pantocin B	−	+/−	+	+
16	−	−	+	+
17	−	−	+	+
18	−	−	+	+
19	−	−	+	+
20	−	−	+	+

[a] The + indicates antibiotic activity. The − indicates no antibiotic activity.

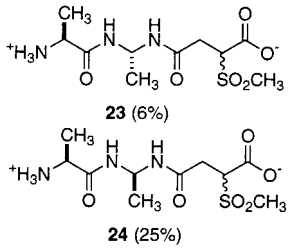
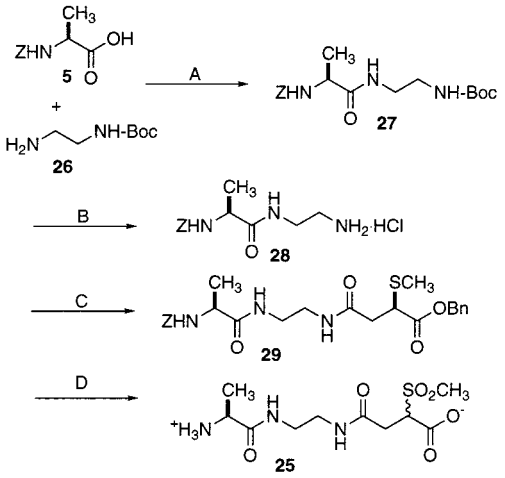


Figure 5. Substituted methylene analogues.

Scheme 2[a]



[a] (A) CDI, THF, 61%. (B) dil HCl, MeOH, 91%. (C) **13a**, EDAC, HOBT, *N*-methylmorpholine, CH_2Cl_2, 77%. (D) i) mCPBA, ii) Pd-black, 5% HCOOH in MeOH, 34%.

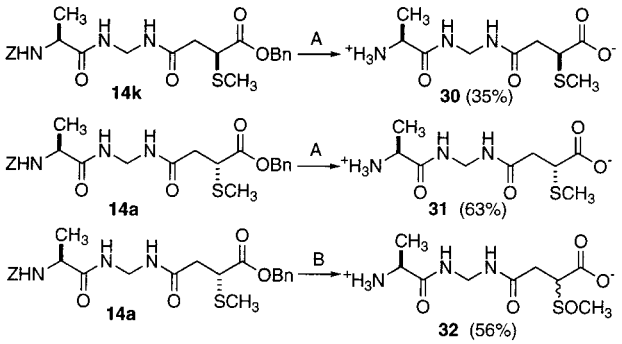
therefore allows it to interact with both pantocin B and **32** as they both possess an isosteric replacement for the *N*-acetyl group present in **4**. The same study noted that aminotransferases could serve as antibiotic targets, and the mechanism of pantocin B provides modest experimental support.[6,15] The lack of activity of the thioether (**30**, **31**) and acrylic acid (**38**, **41**) analogues most likely results from their inability to bind with Acorn due to their lack of an acetyl group isostere.

Table 3. Antibiotic Activity of Mid-Section Analogues against *E. amylovora*[a]

analogue	125 ng
23	0
24	0
25	0

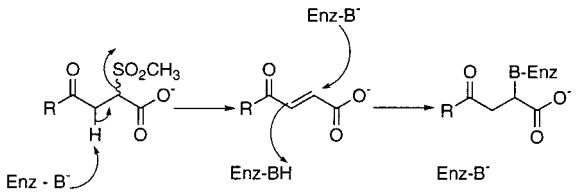
[a] Kill zones are given in mm.

Scheme 3[a]



[a] (A) Pd-black, 5% HCOOH in MeOH. (B) i) 1 equiv of mCPBA, ii) Pd-black, 5% HCOOH in MeOH.

Scheme 4



Conclusions

The synthetic studies described above give a preliminary indication of the roles of the various parts of pantocin B in conveying antibiotic activity. The N-terminal fragment is essential for cellular import but not for interacting with the intracellular target, *N*-acetylornithine aminotransferase, and tolerates a wide variety of substitution as long as the key stereocenter is preserved. The methylenediamine and substituted succinic acid fragments likely interact extensively with the target, and little, if any, structural variation is tolerated. A full explanation of these results will have to await the determination of the three-dimensional structure of *N*-acetylornithine aminotransferase.

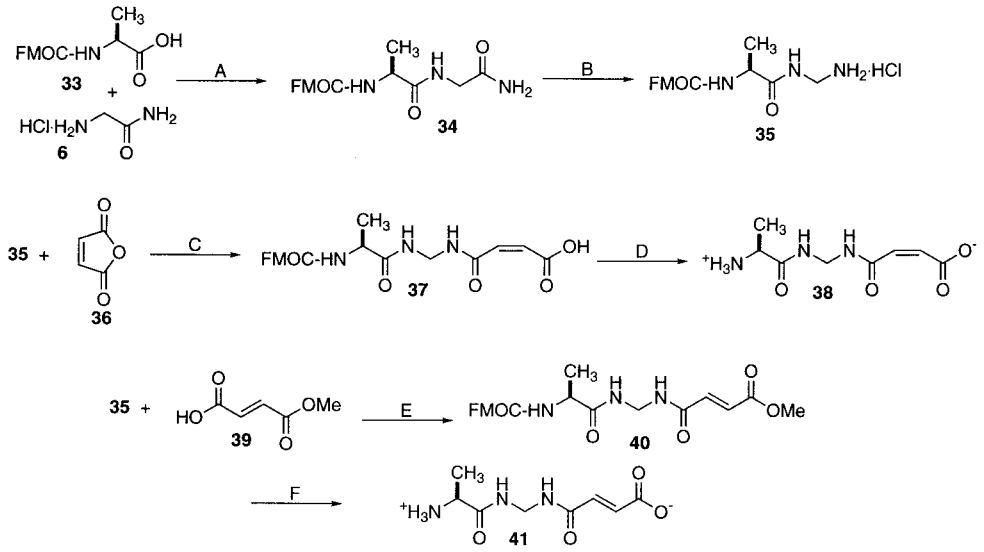
Experimental Section

All reagents and solvents were purchased from commercial suppliers and were used without further purification unless noted. All reagents and solvents were purchased from Aldrich except triflic anhydride (Strem Chemicals), 68% *m*-chloroperbenzoic acid, *Z*-D-alanine, Fmoc-L-alanine, and D-isoleucine (Sigma). D-Alanamide hydrochloride was purchased from Novabiochem. The *Z*-D-isoleucine[16] and methyl hydrogen fumerate[17] were prepared according to standard procedures. Triethylamine was distilled from potassium hydroxide and was stored over potassium hydroxide. The [*I*,*I*-bis(trifluoroacetoxy)iodo]benzene was recrystallized from cold trifluoroacetic acid as needed (clean material is white in color). ^{1}H NMR spectra were recorded at 400 or 500 MHz on a Varian Inova 400 or a Varian Unity 500, respectively. All proton spectra were referenced to residual solvent: 3.30 ppm for CD_3OD, 2.50 ppm for DMSO-d_6, 7.27 ppm for $CDCl_3$, and 4.80 ppm

(16) Bodanszky, M.; Bodanszky, A. *The Practice of Peptide Synthesis*; Springer-Verlag: Berlin, 1984.

(17) Greene, T. W.; Wuts, P. G. M. *Protective Groups in Organic Synthesis*; John Wiley & Sons: New York, 1991.

Scheme 5[a]



[a] (A) EDAC, HOBT, (*i*PR)$_2$NEt, CH_2Cl_2, 50%. (B) i) PIFA, aq MeCN, ii) 1 N HCl, 88%. (C) (*i*Pr)$_2$NEt, CH_2Cl_2, 78%. (D) Et_2NH, DMF, 62%. (E) EDAC, HOBT, (*i*Pr)$_2$NEt, CH_2Cl_2, 70%. (F) i) 0.1 M LiOH, THF, ii) Et_2NH, DMF, 46%.

Table 4. Antibiotic Activity of C-Terminal Analogues against *E. amylovora*[a]

analogue	125 ng	63 ng[b]	31 ng
30	0	0	0
31	0	0	0
32	12	≪	0
38	0	0	0
41	0	0	0

[a] Kill zones are given in mm. [b] < denotes a hazy inhibition zone.

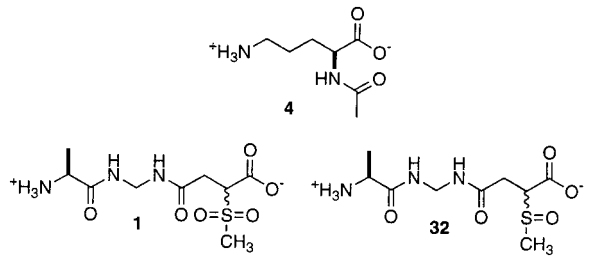


Figure 6. Bioisosteres of *N*-acetylornithine

for D_2O. ^{13}C NMR spectra were recorded at 100 MHz on a Varian VXR-400s spectrometer. Carbon spectra were referenced to residual solvent for DMSO-d_6 (39.51 ppm) and $CDCl_3$ (77.23 ppm). ^{13}C spectra obtained in D_2O were externally referenced to acetonitrile (1.39 ppm) in D_2O. Coupling constants are given in hertz. The phrase "partial data" refers to spectral data of certain compounds that could not be assigned unambiguously because of overlap with other signals. For compounds which exist as diastereomeric mixtures all ^{13}C signals are listed. The *Z*-L-prolyl derivatives exist as pairs of conformational diastereomers due to hindered rotation around the C−N bond.[18] Elemental analyses were run on selected analogues and, while satisfactory, revealed the final zwitterions to typically exist as hydrates. Other general experimental procedures were recently described.[7] The preparation of intermediates **7a**, **8a**, **10a**−**15a** and pantocin B (**1**) have been reported previously.[7]

General Procedure to Synthesize Amide Derivatives (7). In a flame-dried flask under an atmosphere of argon, a *N*-*Z*-protected amino acid (1 equiv) was dissolved in anhydrous tetrahydrofuran. To the resultant solution was added 1,1′-carbonyldiimidazole (1.1 equiv) with stirring. After stirring 1−2 h at room temperature, 1 equiv of the amide hydrochloride and 1 equiv of triethylamine were added to the reaction mixture. The reaction was allowed to proceed at room temperature until judged complete by TLC, typically 1−2 days. The tetrahydrofuran was then removed by evaporation at room temperature under reduced pressure. To the resultant residue was added an aqueous 4% $NaHCO_3$ solution, and the reaction mixture was allowed to stir until a white precipitate was observed. The precipitate was collected by vacuum filtration and dried in vacuo.

***Z*-Glycylglycinamide (7b):** 4.18 g (20.0 mmol) of *Z*-glycine, 2.21 g (20.0 mmol) of glycinamide hydrochloride, 3.31 g (20.2 mmol) of 1,1′-carbonyldiimidazole, 2.60 mL (20.2 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 4.90 g (77%) of a white solid; mp = 178−180 °C. ^1H NMR (500 MHz, DMSO-d_6) δ 3.62 (d, 2 H, J = 6.0 Hz), 3.64 (d, 2 H, J = 6.0 Hz), 5.02 (s, 2 H), 7.08 (s, 1 H), 7.24 (s, 1 H), 7.34 (m, 5 H), 7.49 (t, 1 H, J = 6.0 Hz), 8.06 (m, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 41.81, 43.61, 65.52, 127.72, 127.80, 128.35, 136.97, 156.53, 169.30, 170.85; IR (Nujol mull) 3333, 3194, 3067, 1691, 1646, 1545 cm^{-1}; HRMSFAB$^+$ ($C_{12}H_{16}N_3O_4$, $M^+ + 1$) Calcd: 266.1141 found: 266.1141.

***Z*-L-Phenylalanylglycinamide (7c):** 6.01 g (20.0 mmol) of *Z*-L-phenylalanine, 2.22 g (20.0 mmol) of glycinamide hydrochloride, 3.32 g (20.2 mmol) of 1,1′-carbonyldiimidazole, 2.60 mL (20.2 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 6.43 g (90%) of a white solid. $[\alpha]^{25}_D$ = −3.2° (c = 0.82, methanol); mp = 137−139 °C. ^1H NMR (500 MHz, DMSO-d_6) δ 2.74 (dd, 1 H, J = 9.8, 3.8 Hz), 3.04 (dd, 1 H, J = 3.8, 9.8 Hz), 3.62 (dd, 1 H, J = 5.5, 11.0 Hz), 3.68 (dd, 1 H, J = 5.5, 11.0 Hz), 4.25 (m, 1 H), 4.94 (ab quart, 2 H, J = 12.5, 4.5 Hz), 7.10 (s, 1 H), 7.28 (m, 11 H), 7.58 (d, 1 H, J = 8.5 Hz), 8.24 (t, 1 H, J = 5.5 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 37.24, 41.91, 56.21, 65.24, 126.21, 127.45, 127.67, 128.02, 128.27, 129.18, 129.47, 138.13, 139.63, 155.94, 170.68, 171.65; IR (Nujol mull) 3378, 3302, 3181, 1691, 1659, 1532, 1221 cm^{-1}; HRMSFAB$^+$ ($C_{19}H_{22}N_3O_4$, $M^+ + 1$) Calcd: 356.1610 found: 356.1609.

***Z*-L-Valylglycinamide (7d):** 1.01 g (4.00 mmol) of *Z*-L-valine, 449 mg (4.00 mmol) of glycinamide hydrochloride, 748 mg (4.40 mmol) of 1,1′-carbonyldiimidazole, 560 μL (4.40 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 890 mg (72%) of a white solid. $[\alpha]^{25}_D$ = −6.4° (c = 0.91, methanol); mp = 165−168 °C. ^1H NMR (400 MHz, DMSO-d_6) δ 0.85 (t, 6 H, J = 6.8 Hz), 1.98 (sextet, 1 H, J = 6.8 Hz), 3.60 (d of ab quartets, 2 H, J = 16.8, 9.0, 5.8 Hz), 3.84 (dd, 1 H, J = 7.2, 0.8 Hz), 5.03 (ab quart, 2 H, J = 12.4, 4.0 Hz), 7.04 (s, 1 H), 7.19 (s, 1 H), 7.36 (m, 6 H), 8.10 (m, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 18.05, 19.09, 29.82, 41.69, 60.31, 65.35, 127.60, 127.67, 128.22, 136.86, 156.22, 170.63, 171.30; IR (Nujol mull) 3308, 1697, 1633, 1545, 1297, 1240 cm^{-1}; HRMSFAB$^+$ ($C_{15}H_{22}N_3O_4$, $M^+ + 1$) Calcd: 308.1610 found: 308.1609.

(18) Radzicka, A.; Acheson, S. A.; Wolfenden, R. *Bioorg. Chem.* **1992**, *20*, 382−386.

***Z*-L-Isoleucylglycinamide (7e):** 1.98 g (7.50 mmol) of *Z*-L-isoleucine, 854 mg (7.50 mmol) of glycinamide hydrochloride, 1.36 g (8.30 mmol) of 1,1′-carbonyldiimidazole, 1.10 mL (8.30 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 2.16 g (89%) of a white solid. $[\alpha]^{25}_D = -4.7°$ ($c = 0.90$, methanol); mp = 192−194 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.82 (m, 6 H), 1.10 (m, 1 H), 1.42 (m, 1 H), 1.72 (m, 1 H), 3.60 (dd, 1 H, $J = 5.8$, 11.0 Hz), 3.67 (dd, 1 H, $J = 5.8$, 11.0 Hz) 3.87 (t, 1 H, $J = 7.8$ Hz), 5.02 (ab quart, 2 H, $J = 12.4$, 4.0 Hz), 7.04 (s, 1 H), 7.17 (s, 1 H), 7.36 (m, 6 H), 8.11 (t, 1 H, $J = 5.4$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.95, 15.33, 24.36, 41.80, 59.39, 65.45, 127.70, 127.77, 128.31, 136.94, 156.25, 170.72, 171.45; IR (Nujol mull) 3391, 3308, 1684, 1633, 1551, 1044 cm^{-1}; HRMSFAB$^+$ ($C_{16}H_{24}N_3O_4$, $M^+ + 1$) Calcd: 322.1767 found: 322.1770.

***Z*-L-Prolylglycinamide (7f):** 1.02 g (4.00 mmol) of *Z*-L-proline, 444 mg (4.00 mmol) of glycinamide hydrochloride, 738 mg (4.40 mmol) of 1,1′-carbonyldiimidazole, 560 μL (4.40 mmol) of triethylamine, 20 mL of tetrahydrofuran (Note: the total reaction time was approximately a week); yield 644 mg (53%) of a white solid. $[\alpha]^{25}_D = -39.8°$ ($c = 0.96$, methanol); mp = 120−121 °C. ^{1}H NMR (400 MHz, DMSO-d_6,) δ 1.85 (m, 3 H), 2.12 (m, 1 H), 3.42 (m, 2 H), 3.61 (m, 2 H), 4.16 and 4.26 (dd, 1 H, $J = 3.6$, 5.0 Hz), 5.05 (m, 2 H), 7.08 (m, 2 H), 7.37 (m, 5 H), 8.08 and 8.26 (t, 1 H, $J = 5.6$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 23.00, 23.94, 29.84, 31.10, 41.67, 41.93, 46.57, 47.08, 59.40, 60.16, 65.73, 66.04, 126.99, 127.51, 127.79, 128.20, 128.39, 136.95, 136.79, 153.80, 154.36, 170.55, 170.91, 172.04, 172.30; IR (Nujol mull) 3441, 3321, 1703, 1646, 1589, 1532 cm^{-1}; HRMSFAB$^+$ ($C_{15}H_{20}N_3O_4$, $M^+ + 1$) Calcd: 306.1454 found: 306.1452.

***Z*-D-Alanylglycinamide (7 g):** 1.01 g (4.50 mmol) of *Z*-D-alanine, 503 mg (4.50 mmol) of glycinamide hydrochloride, 812 mg (5.00 mmol) of 1,1′-carbonyldiimidazole, 640 μL (5.00 mmol) of triethylamine, 10 mL of tetrahydrofuran; yield 730 mg (58%) of a white solid. $[\alpha]^{25}_D = 9.3°$ ($c = 1.1$, methanol); mp = 100−102 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.21 (d, 3 H, $J = 7.2$ Hz), 3.61 (t, 2 H, $J = 4.8$ Hz), 4.04 (quintet, 1 H, $J = 7.2$ Hz), 5.02 (ab quart, 2 H, $J = 5.8$, 12.8 Hz), 7.08 (s, 1 H), 7.16 (s, 1 H), 7.35 (m, 5 H), 7.53 (d, 1 H, $J = 7.2$ Hz), 8.07 (t, 1 H, $J = 5.6$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.84, 41.92, 65.46, 127.76, 127.79, 128.33, 136.88, 155.86, 170.82, 172.58; IR (Nujol mull) 3289, 1672, 1538, 1247 cm^{-1}; HRMSFAB$^+$ ($C_{13}H_{18}N_3O_4$, $M^+ + 1$) Calcd: 280.1297 found: 280.1296.

***Z*-D-Isoleucylglycinamide (7h):** 500 mg (1.90 mmol) of *Z*-D-isoleucine, 216 mg (1.90 mmol) of glycinamide hydrochloride, 351 mg (2.10 mmol) of 1,1′-carbonyldiimidazole, 270 μL (2.10 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 211 mg (35%) of a white solid. $[\alpha]^{25}_D = 1.7°$ ($c = 0.40$, methanol); mp = 192−193 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.83 (m, 6 H), 1.11 (m, 1 H), 1.42 (m, 1 H), 1.72 (m, 1 H), 3.59 (dd, 1 H, $J = 5.8$, 11.0 Hz), 3.63 (dd, 1 H, $J = 5.8$, 11.0 Hz), 3.86 (t, 1 H, $J = 8.0$ Hz), 5.02 (ab quart, 2 H, $J = 4.0$, 12.8 Hz), 7.07 (s, 1 H), 7.18 (s, 1 H), 7.33 (m, 6 H), 8.13 (br s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.95, 15.34, 24.38, 36.07, 41.80, 42.06, 43.81, 59.39, 65.46, 127.71, 127.78, 128.32, 134.33, 136.94, 156.26, 170.73, 171.46; IR (Nujol mull) 3290, 1697, 1644, 1537, 1242, 1048 cm^{-1}; HRMSFAB$^+$ ($C_{16}H_{24}N_3O_4$, $M^+ + 1$) Calcd: 322.1767 found: 322.1782.

***Z*-L-Alanyl-L-alanamide (7i):** 507 mg (2.20 mmol) of *Z*-L-alanine, 280 mg (2.20 mmol) of L-alanamide hydrochloride, 450 mg (2.60 mmol) of 1,1′-carbonyldiimidazole, 340 μL (2.40 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 318 mg (49%) of a white solid. $[\alpha]^{25}_D = -29.1°$ ($c = 0.40$, methanol); mp = 200−201 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.19 (d, 6 H, $J = 7.1$ Hz), 4.03 (quintet, 1 H, $J = 7.1$ Hz), 4.18 (quintet, 1 H, $J = 7.1$ Hz), 5.02 (s, 2 H), 6.99 (br s, 1 H), 7.27−7.36 (m, 5 H), 7.47 (d, 1 H, $J = 7.6$ Hz), 7.83 (d, 1 H, $J = 7.6$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 18.02, 18.42, 47.84, 65.35, 127.66, 127.74, 128.31, 136.98, 155.70, 171.93, 174.04; IR (Nujol mull) 3377, 3281, 1677, 1634, 1563, 1240 cm^{-1}; HRMSFAB$^+$ ($C_{14}H_{19}N_3O_4$, $M^+ + 1$) Calcd: 294.1454 found: 294.1453.

***Z*-L-Alanyl-D-alanamide(7j):** 507 mg (2.20 mmol) of *Z*-L-alanine, 290 mg (2.20 mmol) of D-alanamide hydrochloride, 430 mg (2.60 mmol) of 1,1′-carbonyldiimidazole, 340 μL (2.40 mmol) of triethylamine, 20 mL of tetrahydrofuran; yield 325 mg (50%) of a white solid. $[\alpha]^{25}_D = -6.4°$ ($c = 0.50$, methanol); mp = 205−206 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.19 (m, 6 H), 4.04 (quintet, 1 H, $J = 7.3$ Hz), 4.17 (quintet, 1 H, $J = 7.3$ Hz), 5.01 (s, 2 H), 7.05 (br s, 1 H), 7.24 (br s, 1 H), 7.30−7.36 (m, 6 H), 7.51 (d, 1 H, $J = 7.0$ Hz), 8.01 (d, 1 H, $J = 7.0$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.86, 18.19, 47.88, 50.12, 65.40, 127.73, 127.78, 128.33, 136.92, 155.78, 172.00, 174.04; IR (Nujol mull) 3396, 3292, 3189, 1680, 1624, 1537, 1068 cm^{-1}; HRMSFAB$^+$ ($C_{14}H_{19}N_3O_4$, $M^+ + 1$) Calcd: 294.1454 found: 294.1453.

General Procedure to Synthesize Geminal Amino Amide Hydrochlorides (8). Following Loudon's procedure,[9] 1 equiv of [*I,I*-bis-(trifluoroacetoxy)iodo]benzene (PIFA) was dissolved in acetonitrile. To this solution an equal volume of purified (Barnstead, Easy Pure Rf water filtration system) deionized water was added. Finally **7** (1 equiv) was added, and the reaction mixture was allowed to stir at room temperature for approximately 12 h. The reaction mixture was diluted with 1 N HCl (20 equiv) and was washed twice with ether. The aqueous layer was concentrated at reduced pressure. The resultant residue was crystallized from methanol:ether. The resultant white solid was collected by vacuum filtration and was dried in vacuo.

[(Aminomethyl-carbamoyl)methyl]carbamic acid benzyl ester hydrochloride (8b): 308 mg (1.10 mmol) of **7b**, 480 mg (1.10 mmol) of PIFA, 1.5 mL of acetonitrile, 1.5 mL of H_2O; yield 227 mg (71%) of a white solid; mp = 133−135 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.68 (d, 2 H, $J = 6.4$ Hz), 4.22 (d, 2 H, $J = 6.4$ Hz), 5.04 (s, 2 H), 7.36 (m, 5 H), 7.60 (t, 1 H, $J = 6.0$ Hz), 8.20 (br s, 3 H), 8.89 (t, 1 H, $J = 6.0$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 43.32, 44.34, 65.57, 127.14, 127.83, 128.36, 136.93, 156.49, 170.78; IR (Nujol mull) 3302, 1691, 1538, 1291, 1164 cm^{-1}; HRMSFAB$^+$ ($C_{11}H_{16}N_3O_3$, $M^+ - Cl$) Calcd: 238.1192 found: 238.1189.

[1-*S*-(Aminomethyl-carbamoyl)-2-phenylethyl)]carbamic acid benzyl ester hydrochloride (8c): 6.41 g (18.0 mmol) of **7c**, 7.74 g (18.0 mmol) of PIFA, 27 mL of acetonitrile, 27 mL of H_2O; yield 4.08 g (62%) of a white solid. $[\alpha]^{25}_D = -3.9°$ ($c = 0.82$, methanol); mp = 156−159 °C. ^{1}H NMR (500 MHz, DMSO-d_6) δ 2.75 (dd, 1 H, $J = 8.4$, 2.4 Hz), 3.02 (dd, 1 H, $J = 8.4$, 2.4 Hz), 4.29 (m, 3 H), 4.93 (q, 2 H, $J = 10.0$ Hz), 7.21−7.34 (m, 10 H), 7.70 (d, 1 H, $J = 6.8$ Hz), 8.15 (br s, 3 H), 9.13 (t, 1 H, $J = 5.2$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 37.07, 55.94, 56.03, 65.26, 119.09, 126.36, 127.50, 128.23, 129.22, 134.04, 136.89, 137.95, 155.87, 173.17; IR (Nujol mull) 3314, 1697, 1691, 1538, 1266, 1069 cm^{-1}; HRMSFAB$^+$ ($C_{18}H_{22}N_3O_3$, $M^+ - Cl$) Calcd: 328.1661 found: 328.1660.

[1-*S*-(Aminomethyl-carbamoyl)-2-methylpropyl]carbamic acid benzyl ester hydrochloride (8d): 797 mg (2.60 mmol) of **7d**, 1.20 g (2.60 mmol) of PIFA, 6 mL of acetonitrile, 6 mL of H_2O; yield 444 mg (54%) of a white solid. $[\alpha]^{20}_D = -3.5°$ ($c = 0.78$, methanol); mp = 158−160 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.86 (t, 6 H, $J = 6.8$ Hz), 2.00 (sextet, 1 H, $J = 6.8$ Hz), 3.93 (t, 1 H, $J = 7.8$ Hz), 4.20 (m, 2 H), 5.03 (ab quart, 2 H, $J = 11.2$, 12.6 Hz), 7.33 (m, 5 H), 7.49 (d, 1 H, $J = 8.8$ Hz), 8.15 (br s, 3 H), 8.97 (t, 1 H, $J = 6.2$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.97, 19.23, 30.11, 44.46, 60.03, 65.47, 119.10, 127.70, 127.78, 128.33, 136.97, 156.15, 172.82; IR (Nujol mull) 3295, 1697, 1545, 1240, 1050 cm^{-1}; HRMSFAB$^+$ ($C_{14}H_{22}N_3O_3$, $M^+ - Cl$) Calcd: 280.1661 found: 280.1662.

[1-*S*-(Aminomethyl-carbamoyl)-2-methylbutyl]carbamic acid benzyl ester hydrochloride (8e): 704 mg (2.20 mmol) of **7e**, 938 mg (2.20 mmol) of PIFA, 10 mL of acetonitrile, 10 mL of H_2O; yield 366 mg (50%) of a white solid. $[\alpha]^{25}_D = -3.0°$ ($c = 0.84$, methanol); mp = 180 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.83 (m, 6 H), 1.14 (m, 1 H), 1.42 (m, 1 H), 1.75 (m, 1 H), 3.95 (t, 1 H, $J = 8.2$ Hz), 4.21 (m, 2 H), 5.02 (ab quart, 2 H, $J = 11.2$, 12.6 Hz), 7.33 (m, 5 H), 7.49 (d, 1 H, $J = 8.8$ Hz), 8.15 (br s, 3 H), 8.97 (t, 1 H, $J = 6.2$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.86, 15.31, 24.20, 36.10, 38.47, 38.69, 44.63, 58.96, 65.47, 127.71, 127.80, 128.32, 136.91, 156.07, 173.04; IR (Nujol mull) 3283, 1691, 1703, 1551, 1285, 1037 cm^{-1}; HRMSFAB$^+$ ($C_{15}H_{24}N_3O_3$, $M^+ - Cl$) Calcd: 294.1818 found: 294.1820.

[2-*S*-(Aminomethyl-carbamoyl)pyrrolidine]-1-carbamic acid benzyl ester hydrochloride (8f): 303 mg (0.980 mmol) of **7f**, 435 mg (0.980 mmol) of PIFA, 3 mL of acetonitrile, 3 mL of H_2O; yield 203 mg (65%) of a white solid. $[\alpha]^{20}_D = -50.6°$ ($c = 1.1$, methanol); mp = 130−132 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.86 (m, 3 H), 2.15 (m, 1 H), 3.45 (m, 2 H), 4.23 (m, 3 H), 5.05 (m, 2 H), 7.35 (m, 6 H), 8.09 (br s, 3 H), 8.91 (m, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 23.43, 24.46, 30.28, 31.30, 44.90, 45.02, 47.00, 47.54, 59.77, 60.29, 66.34, 66.49, 127.62, 127.95, 128.09, 128.29, 128.76, 128.88, 137.33, 137.43, 154.22, 154.63, 173.77, 174.02; IR (Nujol mull) 3346, 3276, 2052, 1691, 1545, 1367, 1240 cm^{-1}; HRMSFAB$^+$ ($C_{14}H_{20}N_3O_3$, M$^+$− Cl) Calcd: 278.1505 found: 278.1506.

[1-*R*-(Aminomethylcarbamoyl)ethyl]carbamic acid benzyl ester hydrochloride (8g): 179 mg (0.640 mmol) of **7g**, 275 mg (0.640 mmol) of PIFA, 1 mL of acetonitrile, 1 mL of H_2O; yield 117 mg (63%) of a white solid. $[\alpha]^{25}_D = 13.3°$ ($c = 1.32$, methanol); mp = 183 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.23 (d, 3 H, $J = 7.2$ Hz), 4.09 (quintet, 1 H, $J = 7.2$ Hz), 4.17 (dd, 1 H, $J = 6.4$, 5.8 Hz), 4.24 (dd, 1 H, $J = 6.4$, 5.8 Hz), 5.02 (ab quart, 2 H, $J = 12.6$, 14.8 Hz), 7.34 (m, 5 H), 7.61 (d, 1 H, $J = 7.6$ Hz), 8.12 (br s, 3 H), 8.86 (t, 1 H, $J = 6.8$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.82, 45.54, 49.93, 65.47, 127.79, 128.33, 136.89, 155.67, 173.96; IR (Nujol mull) 3340, 1684, 1538, 1297, 1247, 1037 cm^{-1}; HRMSFAB$^+$ ($C_{12}H_{18}N_3O_3$, M$^+$ − Cl) Calcd: 252.1348 found: 252.1348.

[1-*R*-Aminomethyl-carbamoyl)-2-methylbutyl]carbamic acid benzyl ester hydrochloride (8h): 201 mg (0.620 mmol) of **7h**, 249 mg (0.620 mmol) of PIFA, 4 mL of acetonitrile, 4 mL of H_2O; yield 97 mg (46%) of a white solid. $[\alpha]^{25}_D = 1.7°$ ($c = 0.36$, methanol); mp = 183 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.83 (m, 6 H), 1.14 (m, 1 H), 1.42 (m, 1 H), 1.74 (m, 1 H), 3.95 (t, 1 H, $J = 8.0$ Hz), 4.21 (m, 2 H), 5.02 (q, 2 H, $J = 12.3$ Hz), 7.33 (m, 5 H), 7.48 (d, 1 H, $J = 8.8$ Hz), 8.11 (br s, 3 H), 8.95 (t, 1 H, $J = 6.0$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.87, 15.31, 24.19, 36.12, 44.59, 58.89, 65.47, 127.70, 127.79, 128.32, 136.92, 156.05, 173.00; IR (Nujol mull) 3277, 1697, 1670, 1537, 1041 cm^{-1}; HRMSFAB$^+$ ($C_{15}H_{24}N_3O_3$, M$^+$ − Cl) Calcd: 294.1818 found: 294.1821.

[1-*S*-(1-*S*-aminoethylcarbamoyl)ethyl]carbamic acid benzyl ester hydrochloride (8i): 205 mg (0.680 mmol) of **7i**, 302 mg (0.680 mmol) of PIFA, 3 mL of acetonitrile, 3 mL of H_2O; yield 152 mg (72%) of a white solid; mp = 112 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.22 (d, 3 H, $J = 7.2$ Hz), 1.38 (d, 3 H, $J = 6.4$ Hz), 4.08 (quintet, 1 H, $J = 7.2$ Hz), 5.03 (m, 3 H), 7.31−7.37 (m, 5 H), 7.56 (d, 1 H, $J = 7.6$ Hz), 8.18 (br s, 3 H), 8.79 (d, 1 H, $J = 8.0$ Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.86, 18.25, 49.64, 53.61, 65.42, 127.78, 128.33, 136.92, 155.66, 173.15; IR (Nujol mull) br 3200−2500, br 1740−1600, 1239, 1134, 1081 cm^{-1}; HRMSFAB$^+$ ($C_{13}H_{20}N_3O_3$, M$^+$ − Cl) Calcd: 266.1505 found: 266.1505.

[1-*S*-(1-*R*-Aminoethylcarbamoyl)ethyl]carbamic acid benzyl ester hydrochloride (8j): 296 mg (1.00 mmol) of **7j**, 430 mg (1.00 mmol) of PIFA, 4 mL of acetonitrile, 4 mL of H_2O; yield 270 mg (89%) of a sticky glass (NOTE: The aqueous layer was concentrated in vacuo, and the material was used directly since it failed to crystallize); ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.22 (d, 3 H, $J = 6.8$ Hz), 1.36 (d, 3 H, $J = 6.4$ Hz), 4.09 (quintet, 1 H, $J = 6.8$ Hz), 4.89 (br s, 1 H), 5.02 (m, 2 H), 7.31−7.37 (m, 5 H), 7.57 (d, 1 H, $J = 7.2$ Hz), 8.11 (br s, 3 H), 8.80 (s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 18.08, 18.20, 49.86, 54.01, 65.40, 127.76, 127.82, 128.34, 136.93, 155.57, 173.48; IR ($CHCl_3$) br 3400−2500, 1684, 1526, 1221 cm^{-1}; HRMSFAB$^+$ ($C_{13}H_{20}N_3O_3$, M$^+$ − Cl) Calcd: 266.1505 found: 266.1505.

General Procedure to Synthesize 2-Hydroxysuccinic acid-1-benzyl Ester (10). Following Miller's procedure,[19] 2.4 equiv of trifluoroacetic anhydride was placed in a dried flask under an argon atmosphere, and the flask was placed in an ice bath. To the chilled anhydride, malic acid (1 equiv) was added, and the resultant suspension was stirred at 0 °C until all the malic acid was dissolved. The clear solution was then concentrated on a rotary evaporator not exceeding a water bath temperature of 30 °C. To the resultant solid residue benzyl alcohol was added, eventually giving rise to a clear solution. The reaction mixture was stirred overnight at room temperature, diluted with ethyl acetate, and extracted with an aqueous 4% $NaHCO_3$ solution. The combined aqueous extracts were washed with ethyl acetate and acidified to pH = 2 with concentrated HCl. The acidified solution was extracted with ethyl acetate. The combined organic extracts were dried over $MgSO_4$ and concentrated in vacuo to give a clear oil.

2-*R*-Hydroxysuccinic acid 1-benzylester (10b): 3.98 g (30.0 mmol) of D-malic acid, 10.0 mL (72.0 mmol) of trifluoroacetic anhydride, 10 mL of benzyl alcohol; yield 5.27 g (86%) of a clear oil. $[\alpha]^{25}_D = 16.7°$ ($c = 1.60$, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.86 (dd, 1 H, $J = 6.0$, 16.8 Hz), 2.93 (dd, 1 H, $J = 4.4$, 16.8 Hz), 4.55 (dd, 1 H, $J = 4.4$, 6.0 Hz), 5.24 (s, 2 H), 7.37 (m, 5 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.55, 67.25, 68.15, 128.67, 128.91, 134.99, 173.23, 176.13; IR (neat) br 3480−2660, 1722, 1399, 1221, 1101 cm^{-1}; HRMSFAB$^+$ ($C_{11}H_{13}O_5$, M$^+$ + 1) Calcd: 225.0763 found: 225.0762.

General Procedure to Synthesize 2-Hydroxysuccinic Acid-1-benzyl Ester-4(4-methoxybenzyl) Ester (11). In a dried flask under an atmosphere of argon **10** (1 equiv) was dissolved in anhydrous dimethylformamide. To the resultant solution was added potassium bicarbonate (K_2CO_3, 2 equiv) with vigorous stirring. 4-methoxybenzyl chloride was added to the stirred suspension, and the reaction mixture was stirred at room temperature for 4 days. The reaction mixture was diluted with ethyl acetate, and the organics were washed three times with dIH_2O. The organics were dried over $MgSO_4$ and concentrated at reduced pressure resulting in a viscous oil. After flash chromatography on Si gel (70:30 hexanes:ethyl acetate) the desired diester **11** was isolated as a clear oil.

2-*R*-Hydroxysuccinic acid-1-benzylester-4-(4-methoxybenzyl) ester (11b): 4.09 g (18.0 mmol) of **10b**, 2.90 mL (20.0 mmol) of 4-methoxybenzyl chloride, 5.01 g (36.0 mmol) of K_2CO_3, 36 mL of dimethylformamide; yield 3.66 g (58%) of a clear oil. $[\alpha]^{25}_D = 18.7°$ ($c = 1.22$, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.83 (dd, 1 H, $J = 6.0$, 16.4 Hz), 2.89 (dd, 1 H, $J = 4.4$, 16.4 Hz), 3.20 (br s, 1 H), 3.81 (s, 3 H), 4.54 (br s, 1 H), 5.05 (s, 2 H), 5.19 (s, 2 H), 6.88 (d, 2 H, $J = 8.4$ Hz), 7.26 (d, 2 H, $J = 8.4$ Hz), 7.34 (m, 5 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.91, 55.50, 66.86, 67.92, 114.16, 127.71, 128.61, 128.82, 128.85, 130.45, 135.15, 159.94, 170.50, 173.39; IR (neat) 3486, 2959, 1748, 1614, 1519, 1215 cm^{-1}; HRMSEI$^+$ ($C_{19}H_{20}O_6$, M$^+$) Calcd: 344.1260 found: 344.1258

General Procedure to Synthesize 2-Methanesulfanyl Succinic Acid 1-Benzyl Ester-4-(4-methoxybenzyl) Ester (12). In a flame-dried Schlenck flask under an argon atmosphere **11** (1 equiv) was dissolved in anhydrous dichloromethane. The resultant solution was chilled to −78 °C in a dry ice/acetone bath. To the chilled solution was added 2,6-lutidine (1.3 equiv) with stirring. Finally triflic anhydride (1.2 equiv) was added dropwise to the reaction mixture. After complete addition of the triflic anhydride, the reaction mixture was stirred for 4 h at −78 °C. In a second flame-dried flask 15-crown-5 (4 equiv) was dissolved in anhydrous dimethylformamide. Sodium thiomethoxide (4 equiv) was added to the solution of crown ether and was stirred vigorously. The resultant suspension was added in small portions over 1 h via an addition funnel to the stirred triflate solution under positive argon flow. (*Note: The reaction mixture must be stirred vigorously to ensure adequate mixing*.) When the suspension was completely added to the triflate solution, the reaction mixture was allowed to warm slowly to room temperature. The progress of the reaction was monitored by ^{1}H NMR of quenched (1 N HCl) aliquots; the reaction was judged complete when no chlorosuccinate derivative was observed in the ^{1}H NMR. The reaction mixture was diluted with ether and washed with dIH_2O and 1 N HCl. The organics were dried over $MgSO_4$ and concentrated at reduced pressure, yielding a crude oil. The ether was chased with hexanes on the rotary evaporator, and the resultant white solid was recrystallized from ether:hexanes to give **12**. The solid was collected by vacuum filtration and dried in vacuo.

2-S-Methanesulfanyl succinic acid-1-benzyl ester-4-(4-methoxybenzyl) ester (12b): 3.03 g (8.70 mmol) of **11b**, 1.80 mL (11.0 mmol) of triflic anhydride, 1.40 mL (12.0 mmol) of 2,6-lutidine, 25 mL of dichloromethane, 2.54 g (36.0 mmol) of sodium thiomethoxide, 7.00 mL (36.0 mmol) of 15-crown-5, 30 mL of dimethylformamide; yield 2.51 g (76%) of a white solid. $[\alpha]^{25}_D = -18.2°$ ($c = 0.95$, $CHCl_3$); mp = 49−51 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.12 (s, 3 H), 2.72 (dd,

(19) Miller, M. J.; Bajwa, J. S.; Mattingly, P. G.; Peterson, K. *J. Org. Chem.* **1982**, *47*, 49284933.

1 H, J = 5.6, 17.0 Hz), 3.06 (dd, 1 H, J = 9.6, 17.0 Hz), 3.67 (dd, 1 H, J = 5.6, 9.6 Hz), 3.81 (s, 3 H), 5.04 (ab quart, 2 H, J = 3.6, 12.0 Hz), 5.15 (ab quart, 2 H, J = 12.4, 20.0 Hz), 6.88 (d, 2 H, J = 8.8 Hz), 7.26 (d, 2 H, J = 8.8 Hz), 7.36 (m, 5 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.14, 36.21, 42.45, 55.50, 66.85, 67.22, 114.15, 127.79, 128.31, 128.49, 128.75, 130.37, 135.82, 159.92, 170.64, 171.22; IR (Nujol mull) 1722, 1614, 1513, 1240, 1151 cm^{-1}; HRMSEI$^+$ ($C_{20}H_{22}O_5S$, M^+) Calcd: 374.1188 found: 374.1189.

General Procedure to Synthesize 2-Methanesulfanyl-succinic acid 1-benzyl ester (13). A flask containing 10% trifluoroacetic acid in dichloromethane solution was chilled in an icebath. **12** (0.1 g/mL) was dissolved in the chilled acid solution, and the resultant solution was stirred for 3−4 h while letting the reaction temperature warm slowly to room temperature. The reaction mixture was concentrated on a rotary evaporator, and the residue was dissolved in ether. The ether solution was extracted with an aqueous 4% $NaHCO_3$ solution. The combined aqueous extracts were acidified to pH = 1 with concentrated HCl and extracted with ethyl acetate. The combined ethyl acetate extracts were dried over $MgSO_4$ and concentrated on a rotary evaporator, and the resultant clear oil was dried in vacuo.

2-*S*-Methanesulfanyl succinic acid 1-benzyl ester (13b): 2.06 g (5.30 mmol) of **12b**, 20 mL of 10% trifluoroacetic acid in dichloromethane; yield 1.51 g (quantitative yield) of a clear oil. $[\alpha]^{25}_D$ = −27.3° (c = 1.30, $CHCl_3$);[20] ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.14 (s, 3 H), 2.76 (dd, 1 H, J = 5.6, 17.2 Hz), 3.12 (dd, 1 H, J = 10.0, 17.2 Hz), 3.66 (dd, 1 H, J = 5.6, 10.00 Hz), 5.22 (ab quart, 2 H, J = 5.2, 12.8 Hz), 7.36 (m, 5 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.18, 35.80, 42.09, 67.40, 128.32, 128.55, 128.79, 135.69, 171.14, 176.36; IR (neat) br 3400−2400, 1805, 1722, 1152 cm^{-1}; HRMSEI$^+$ ($C_{12}H_{14}O_4S$, M^+) Calcd: 254.0613 found: 254.0607.

General Procedure to Synthesize Coupling Product (14). In a dried flask under argon, **8** (1 equiv) was suspended in anhydrous dichloromethane. To the suspension *N*-ethylmorpholine (1.5 equiv) was added. The reaction mixture was stirred for 10 min at room temperature and then chilled to 0−5 °C. To the chilled suspension, a solution of **13** (1 equiv) in anhydrous dichloromethane was added. *N*-hydroxybenzotriazole hydrate (HOBT, 1.3 equiv) was added to the chilled reaction mixture with stirring. Finally *N*-ethyl-*N*′-[(3-dimethylamino) propyl]-carbodiimide (EDAC, 1.2 equiv) was added to the reaction mixture. The reaction temperature was maintained at 0 °C for 1 h before warming slowly to room temperature. Upon completion the dichloromethane was removed under partial vacuum. To the resultant residue 1 N HCl was added, and the white precipitate was collected by vacuum filtration and dried in vacuo.

***N*-[(2-Benzyloxycarbonylamino-acetyl amino)methyl] 2-*R*-methanesulfanyl succinamic acid benzyl ester (14b):** 504 mg (1.80 mmol) of **8b**, 30 mL of dichloromethane, 459 mg (1.80 mmol) of **13a**, 10 mL of dichloromethane, 418 mg (2.20 mmol) of EDAC, 355 mg (2.30 mmol) of HOBT, 340 μL (2.70 mmol) of *N*-ethylmorpholine; yield 540 mg (62%) of a white solid; mp = 82−83 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 2.06 (s, 3 H), 2.74 (dd, 1 H, J = 10.0, 6.0 Hz), 3.58 (d, 2 H, J = 6.0 Hz), 3.64 (dd, 1 H, J = 6.0, 10.0 Hz), 4.38 (t, 2 H, J = 6.0 Hz), 5.02 (s, 2 H), 5.13 (ab quart, 2 H, J = 12.6, 15.2 Hz), 7.35 (m, 11 H), 8.47 (m, 1 H), 8.60 (m, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.43, 36.23, 42.17, 43.18, 65.41, 65.85, 127.66, 127.70, 127.74, 127.96, 128.30, 128.37, 136.02, 137.02, 156.40, 169.54, 170.90; IR (Nujol mull) 3302, 1716, 1646, 1545 cm^{-1}; HRMSFAB$^+$ ($C_{23}H_{28}N_3O_6S$, M^+ + 1) Calcd: 474.1699 found: 474.1699.

***N*-((2-*S*-Benzyloxycarbonylamino-3-phenyl-propanoylamino)-methyl] 2-*R*-methanesulfanyl succinamic acid benzyl ester (14c):** 435 mg (1.20 mmol) of **8c**, 18 mL of dichloromethane, 299 mg (1.20 mmol) of **13a**, 6 mL of dichloromethane, 279 mg (1.40 mmol) of EDAC, 249 mg (1.60 mmol) of HOBT, 220 μL (1.8 mmol) of *N*-ethylmorpholine; yield 391 mg (59%) of a white solid; mp = 126−128 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 2.06 (s, 3 H), 2.70 (dd, 1 H, J = 10.8, 2.8 Hz), 2.76 (dd, 1 H, J = 9.6, 6.0 Hz), 2.94 (dd, 1 H, J = 4.0, 9.6 Hz), 3.64 (dd, 1 H, J = 6.0, 9.6 Hz), 4.23 (m, 1 H), 4.40 (m, 2 H), 4.91 (ab quart, 2 H, J = 13.0, 3.2 Hz), 5.13 (m, 2 H), 7.10−7.44 (m, 16 H), 8.63 (t, 1 H, J = 6.0 Hz), 8.68 (t, 1 H, J = 6.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.41, 30.67, 36.26, 37.53, 42.21, 43.28, 56.02, 65.16, 65.89, 126.21, 127.42, 127.72, 127.97, 128.23, 129.20, 136.04, 137.00, 138.06, 155.76, 169.62, 170.92, 172.58; IR (Nujol mull) 3295, 1729, 1705, 1652, 1538 cm^{-1}; HRMSFAB$^+$ ($C_{30}H_{34}N_3O_6S$, M^+ + 1) Calcd: 564.2168 found: 564.2170.

***N*-[(2-*S*-Benzyloxycarbonylamino-3-methylbutrylamino)methyl] 2-*R*-methanesulfanyl succinamic benzyl ester (14d):** 608 mg (1.90 mmol) of **8d**, 35 mL of dichloromethane, 481 mg (1.90 mmol) of **13a**, 5 mL of dichloromethane, 439 mg (2.30 mmol) of EDAC, 388 g (2.50 mmol) of HOBT, 360 μL (2.90 mmol) of *N*-ethylmorpholine; yield 708 mg (73%) of a white solid; mp = 180 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 0.82 (t, 6 H, J = 6.4 Hz), 1.91 (sextet, 1 H, J = 6.4 Hz), 2.05 (s, 3 H), 2.73 (dd, 1 H, J = 9.2, 6.4 Hz), 3.62 (dd, 1 H, J = 9.2, 6.0 Hz), 3.80 (t, 1 H, J = 8.0 Hz), 4.37 (q, 2 H, J = 5.6 Hz), 5.01 (ab quart, 2 H, J = 12.8, 4.0 Hz), 5.12 (q, 2 H, J = 12.8 Hz), 7.21 (d, 1 H, J = 8.8 Hz), 7.34 (m, 10 H), 8.50 (t, 1 H, J = 5.7 Hz), 8.63 (t, 1 H, J = 5.7 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.38, 18.09, 19.13, 30.35, 36.19, 42.20, 43.13, 59.96, 65.35, 65.86, 127.72, 127.98, 128.31, 128.38, 136.02, 137.05, 156.02, 169.53, 170.89, 171.74; IR (Nujol mull) 3302, 1735, 1684, 1640 cm^{-1}; HRMS ($C_{26}H_{34}N_3O_6S$, M^+ + 1) Calcd: 516.2168 found: 516.2172.

***N*-[(2-*S*-Benzyloxycarbonylamino-3-methylpentanoylamino)methyl] 2-*R*-methanesulfanyl succinamic benzyl ester (14e):** 330 mg (1.00 mmol) of **8e**, 15 mL of dichloromethane, 258 mg (1.00 mmol) of **13a**, 5 mL of dichloromethane, 256 mg (1.20 mmol) of EDAC, 213 mg (1.30 mmol) of HOBT, 190 μL (1.50 mmol) of *N*-ethylmorpholine; yield 314 mg (60%) of a white solid; mp = 167−168 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 0.78 (m, 6 H), 1.07 (m, 1 H), 1.38 (m, 1 H), 1.66 (m, 1 H), 2.05 (s, 3 H), 2.74 (dd, 1 H, J = 9.4, 5.2 Hz), 3.62 (dd, 1 H, J = 6.0, 9.4 Hz), 3.87 (t, 1 H, J = 8.4 Hz), 4.37 (m, 2 H), 5.01 (ab quart, 2 H, J = 12.4, 4.4 Hz), 5.13 (q, 2 H, J = 12.4 Hz), 7.24 (d, 1 H, J = 9.2 Hz), 7.35 (m, 10 H), 8.51 (t, 1 H, J = 5.8 Hz), 8.63 (t, 1 H, J = 5.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.90, 13.37, 15.26, 24.30, 36.19, 36.49, 42.18, 43.07, 58.90, 65.35, 65.86, 127.66, 127.72, 127.98, 128.29, 128.38, 136.02, 137.04, 155.91, 169.53, 170.88, 171.81; IR (Nujol mull) 3302, 3073, 1741, 1697, 1652, 1532, 1164 cm^{-1}; HRMSFAB$^+$ ($C_{27}H_{36}N_3O_6S$, M^+ + 1) Calcd: 530.2325 found: 530.2331.

2-S-{[(3-Benzyloxycarbonyl-3-*R*-methanesulfanyl-propanoyl-amino)methyl]carbamoyl]pyrrolidine-1-carboxylic acid benzyl ester (14f): 499 mg (1.60 mmol) of **8f**, 20 mL of dichloromethane, 407 mg (1.60 mmol) of **13a**, 10 mL of dichloromethane, 367 mg (1.90 mmol) of EDAC, 329 mg (2.10 mmol) of HOBT, 300 μL (2.40 mmol) of *N*-ethylmorpholine; yield 578 mg (70%). *NOTE*: Change in workup, the reaction mixture was diluted with dichloromethane, and the organics were washed with 1 N HCl and an aqueous 4% $NaHCO_3$ solution. The organics were dried over $MgSO_4$ and concentrated under partial vacuum. The resultant residue was crystallized from dichloromethane:hexanes to give **14f**; mp = 143−144 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.80 (m, 3 H), 2.02 (m, 4 H), 2.70 (m, 2 H), 3.02 (m, 2 H), 4.20 (m, 1 H), 4.38 (m, 2 H), 5.05 (m, 4 H), 7.34 (m, 11 H), 8.40 (m, 2 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.48, 23.89, 24.69, 29.23, 42.79, 47.28, 47.72, 60.95, 67.14, 67.62, 128.23, 128.34, 128.46, 128.74, 135.87, 136.52, 170.63, 171.57; IR (Nujol mull) 3308, 1710, 1703, 1646, 1526 cm^{-1}; HRMSFAB$^+$ ($C_{26}H_{32}N_3O_6S$, M^+ + 1) Calcd: 514.2012 found: 514.2010.

***N*-[(2-*R*-Benzyloxycarbonyl-propanoylamino)methyl] 2-*R*-methanesulfanyl succinamic acid benzyl ester (14 g):** 302 mg (1.00 mmol) of **8g**, 15 mL of dichloromethane, 264 mg (1.00 mmol) of **13a**, 5 mL of dichloromethane, 243 mg (1.30 mmol) of EDAC, 184 mg (1.40 mmol) of HOBT, 200 μL (1.60 mmol) of *N*-ethylmorpholine; yield 298 mg (59%) of a white solid; mp = 152−153 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.16 (d, 3 H, J = 7.2 Hz), 2.05 (s, 3 H), 2.74 (dd, 1 H, J = 9.6, 5.6 Hz), 3.64 (dd, 1 H, J = 9.6, 6.4 Hz), 4.02 (quintet, 1 H, J = 7.2 Hz), 4.37 (m, 2 H), 5.00 (ab quart, 2 H, J = 12.6, 6.8 Hz), 5.13 (ab quart, 2 H, J = 12.8, 14.0 Hz), 7.34 (m, 11 H), 8.44 (t, 1 H, J = 5.6 Hz), 8.58 (t, 1 H, J = 5.6 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.43, 18.16, 36.25, 42.18, 43.31, 49.81, 65.33, 65.86, 127.70, 127.97, 128.31, 136.02, 137.00, 155.54, 169.57, 170.90,

(20) The synthesis of **13a** was originally presented in ref 7 and was shown to be a 5:1 mixture of enantiomers, *R* predominating with and $[\alpha]^{25}_D$ = 27.9° (c = 0.41, $CHCl_3$); correspondingly **13b** exists as a 5:1 mixture of enantiomers with *S* predominating.

172.97; IR (Nujol mull) 3352, 3302, 1710, 1646, 1538 cm^{-1}; HRMS-FAB$^+$ ($C_{24}H_{30}N_3O_6S$, $M^+ + 1$) Calcd: 488.1855 found: 488.1860.

***N*-[(2-*R*-Benzyloxycarbonylamino-3-methylpentanoylamino)methyl] 2-*R*-methanesulfanyl succinamic benzyl ester (14h):** 88 mg (0.27 mmol) of **8h**, 15 mL of dichloromethane, 71 mg (0.27 mmol) of **13a**, 6 mL of dichloromethane, 62 mg (0.32 mmol) of EDAC, 60 mg (0.35 mmol) of HOBT, 50 μL (0.40 mmol) of *N*-ethylmorpholine; yield 93 mg (66%) of a white solid; mp = 164−165 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 0.78 (m, 6 H), 1.07 (m, 1 H), 1.38 (m, 1 H), 1.66 (m, 1 H), 2.05 (s, 3 H), 2.74 (dd, 1 H, J = 6.8, 9.4 Hz), 3.62 (dd, 1 H, J = 5.8, 9.4 Hz), 3.87 (t, 1 H, J = 8.0 Hz), 4.37 (m, 2 H), 5.01 (ab quart, 2 H, J = 12.8, 4.0 Hz), 5.13 (q, 2 H, J = 12.7 Hz), 7.24 (d, 1 H, J = 9.2 Hz), 7.35 (m, 10 H), 8.53 (t, 1 H, J = 5.9 Hz), 8.79 (t, 1 H, J = 5.9 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.89, 13.36, 15.26, 24.29, 36.19, 36.48, 42.18, 43.09, 58.88, 65.35, 65.86, 127.64, 127.70, 127.98, 128.29, 128.38, 136.01, 137.03, 155.90, 169.52, 170.87, 171.79; IR (Nujol mull) 3310, 1731, 1650, 1543, 1162 cm^{-1}; HRMS-FAB$^+$ ($C_{27}H_{36}N_3O_6S$, $M^+ + 1$) Calcd: 530.2326 found: 530.2325.

***N*-[1-*S*-(2-*S*-Benzyloxycarbonylamino-propanoylamino)-ethyl] 2-*R*-methanesulfanyl succinamic acid benzyl ester (14i):** 61 mg (0.20 mmol) of **8i**, 50 mg (0.20 mmol) of **13a**, 5 mL of dichloromethane, 54 mg (0.28 mmol) of EDAC, 50 mg (0.32 mmol) of HOBT, 40 μL (0.30 mmol) of *N*-ethylmorpholine; yield 74 mg (74%) of a white solid (NOTE: Change in workup, the reaction mixture was partitioned between ethyl acetate and 1 N HCl. The organics were washed with a saturated aqueous $NaHCO_3$ solution and dried over $MgSO_4$. After filtration, the organics were concentrated in vacuo, giving **14i** cleanly.); mp = 143−144 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.15 (d, 3 H, J = 7.2 Hz), 1.22 (d, 3 H, J = 6.4 Hz), 2.05 (s, 3 H), 2.70 (dd, 1 H, J = 6.4, 8.8 Hz), 3.60 (dd, 1 H, J = 8.8, 6.0 Hz), 3.99 (m, 1 H), 5.00 (ab quart, 2 H, J = 6.4, 12.2 Hz), 5.12 (ab quart, 2 H, J = 6.0, 13.0 Hz), 5.40 (m, 1 H) 7.20−7.34 (m, 11 H), 8.08 (d, 1 H, J = 7.4 Hz), 8.23 (d, 1 H, J = 7.4 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.42, 18.18, 20.72, 42.35, 49.79, 52.27, 65.30, 65.87, 66.15, 127.69, 127.75, 127.94, 128.04, 128.30, 128.38, 128.47, 136.02, 137.00, 155.57, 168.07, 170.90, 171.36; IR (Nujol mull) 3290, 1722, 1664, 1565, 1144 cm^{-1}; HRMSFAB$^+$ ($C_{25}H_{32}N_3O_6S$, $M^+ + 1$) Calcd: 502.2012 found: 502.2012.

***N*-[1-*R*-(2-*S*-Benzyloxycarbonylamino-propanoylamino)-ethyl] 2-*R*-methanesulfanyl succinamic acid benzyl ester(14j):** 172 mg (0.570 mmol) of **8j**, 155 mg (0.610 mmol) of **13a**, 20 mL of dichloromethane, 156 mg (0.800 mmol) of EDAC, 131 mg (0.860 mmol) of HOBT, 130 μL (1.00 mmol) of *N*-ethylmorpholine; yield 202 mg (71%) of a white solid (NOTE: Change in workup, the reaction mixture was partitioned between ethyl acetate and 1 N HCl. The organics were washed with a saturated aqueous $NaHCO_3$ solution. The organics were dried over $MgSO_4$. After filtration, the reaction was concentrated in vacuo, giving **14j** cleanly.); mp = 123−125 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.17 (m, 6 H), 2.07 (s, 3 H), 2.75 (dd, 1 H, J = 5.6, 10.2 Hz), 3.63 (dd, 1 H, J = 5.0, 10.2 Hz), 4.01 (m, 1 H), 5.00−5.18 (m, 4 H), 5.40 (m, 1 H), 7.32−7.40 (m, 11 H), 8.05 (d, 1 H, J = 7.4 Hz), 8.22 (d, 1 H, J = 7.4 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 14.04, 18.98, 21.16, 36.99, 42.64, 50.34, 52.83, 65.83, 66.34, 128.17, 128.45, 128.52, 128.79, 128.85, 128.97, 136.52, 137.48, 156.00, 168.36, 171.42, 171.86; IR (Nujol mull) 3300, 1728, 1664, 1529, 1259 cm^{-1}; HRMSFAB$^+$ ($C_{25}H_{32}N_3O_6S$, $M^+ + 1$) Calcd: 502.2012 found: 502.2012.

***N*-[(2-*S*-Benzyloxycarbonylamino-propanoylamino)methyl] 2-*S*-methanesulfanyl succinamic acid benzyl ester (14k):** 505 mg (1.70 mmol) of **8a**, 30 mL of dichloromethane, 438 mg (1.70 mmol) of **13b**, 10 mL of dichloromethane, 394 mg (2.00 mmol) of EDAC, 346 mg (2.20 mmol) of HOBT, 330 μL (2.60 mmol) of *N*-ethylmorpholine; yield 523 mg (62%) of a white solid; mp = 141−143 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.16 (d, 3 H, J = 7.2 Hz), 2.06 (s, 3 H), 2.74 (dd, 1 H, J = 10.0, 5.6 Hz), 3.64 (dd, 1 H, J = 6.0, 10.0 Hz), 4.02 (quintet, 1 H, J = 7.2 Hz), 4.37 (m, 2 H), 5.00 (ab quart, 2 H, J = 12.4, 6.8 Hz), 5.13 (ab quart, 2 H, J = 12.4, 14.4 Hz), 7.35 (m, 11 H), 8.44 (t, 1 H, J = 6.0 Hz), 8.58 (t, 1 H, J = 6.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.42, 18.16, 36.25, 42.18, 43.31, 49.80, 65.32, 65.85, 127.69, 127.97, 128.30, 128.38, 136.01, 136.99, 155.54, 169.56, 170.89, 172.69; IR (Nujol mull) 3314, 1710, 1700, 1646, 1532, 1304 cm^{-1}; HRMSFAB$^+$ ($C_{24}H_{30}N_3O_6S$, $M^+ + 1$) Calcd: 488.1855 found: 488.1860.

Scheme A: General Procedure to Synthesize Sulfone (15). In a dried flask under argon, 1 equiv of **14** was suspended in anhydrous dichloromethane. The suspension was chilled to approximately 0 °C. In a second dried flask under argon 2.2 equiv of *m*-chloroperbenzoic acid (mCPBA) was dissolved in anhydrous dichloromethane. The solution of mCPBA was added in portions to the suspension of **14**. The reaction mixture was stirred for 3−4 h at 0 °C. When the oxidation was complete, 2−4 mL of a saturated sodium thiosulfate solution was added to the reaction mixture to quench the residual mCPBA. The reaction mixture was then stirred vigorously for 10−20 min at which time the reaction mixture was partitioned between dIH_2O and chloroform. The organics were washed three times with saturated aqueous $NaHCO_3$ solution and one time with brine. The organics were then dried over $MgSO_4$ and concentrated under partial vacuum. The resultant solid was dried in vacuo to give the sulfone **15** cleanly.

***N*-[(2-*S*-Benzyloxycarbonylamino-3-phenylpropanoylamino)methyl] 2-methanesulfonyl succinamic acid benzyl ester (15c):** 403 mg (0.700 mmol) of **14c**, 15 mL of dichloromethane, 461 mg (1.50 mmol) of mCPBA, 15 mL of dichloromethane; yield 284 mg (68%) of a white solid; mp = 145−147 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.71 (t, 1 H, J = 10.8 Hz), 2.92 (m, 3 H), 3.14 (s, 3 H), 4.23 (m, 1 H), 4.41 (m, 2 H), 4.67 (m, 1 H), 4.92 (ab quart, J = 12.8, 4.4 Hz), 5.21 (m, 2 H), 7.00−7.40 (m, 16 H), 8.71 (q, 1 H, J = 5.8 Hz), 8.80 (q, 1 H, J = 5.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 30.55, 37.53, 43.44, 50.16, 55.98, 64.16, 65.16, 67.15, 126.21, 127.42, 127.64, 127.91, 127.98, 128.26, 128.41, 128.80, 129.21, 130.64, 135.19, 136.98, 138.01, 138.07, 155.76, 165.28, 168.42, 168.48, 172.05; IR (Nujol mull) 3295, 1741, 1697, 1652, 1538 cm^{-1}; HRMSFAB$^+$ ($C_{30}H_{34}N_3O_8S$, $M^+ + 1$) Calcd: 596.2067 found: 596.2075.

***N*-[(2-*S*-Benzyloxycarbonylamino-3-methylbutrylamino)methyl] 2-methanesulfonyl succinamic acid benzyl ester (15d):** 194 mg (0.390 mmol) of **14d**, 10 mL of dichloromethane, 253 mg (0.850 mmol) of mCPBA, 10 mL of dichloromethane; yield 109 mg (53%) of a white solid; mp = 116−117 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.89 (t, 6 H, J = 6.4 Hz), 1.91 (sextet, 1 H, J = 6.4 Hz), 2.78−2.97 (m, 2 H), 3.13 (s, 3 H), 3.85 (m, 1 H), 4.93 (m, 2 H), 4.65 (dd, 2 H, J = 3.6, 10.8 Hz), 5.01 (ab quart, 2 H, J = 12.6, 6.4 Hz), 5.20 (d of ab quarts, 2 H, J = 2.4, 12.4, 6.0 Hz), 7.22 (d, 1 H, J = 8.8 Hz), 7.36 (m, 10 H), 8.52 (t, 1 H, J = 6.0 Hz), 8.79 (t, 1 H, J = 6.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 18.07, 19.12, 30.33, 30.48, 43.28, 59.91, 64.16, 65.37, 67.14, 127.66, 127.95, 128.21, 128.31, 135.18, 137.04, 156.03, 165.24, 168.38, 171.76; IR (Nujol mull) 3302, 1735, 1691, 1640, 1540 cm^{-1}; HRMSFAB$^+$ ($C_{26}H_{34}N_3O_8S$, $M^+ + 1$) Calcd: 548.2067 found: 548.2065.

***N*-[(2-*S*-Benzyloxycarbonyl-3-methylpentanoylamino)methyl] 2-methanesulfonyl succinamic acid benzyl ester (15e):** 252 mg (0.470 mmol) of **14e**, 8 mL of dichloromethane, 314 mg (1.03 mmol) of mCPBA, 8 mL of dichloromethane; yield 246 mg (93%) of a white solid. [Note: a precipitate was observed in the organic layer so the organics were concentrated directly under partial vacuum. The resultant residue was triturated with a dichloromethane: hexanes mixture to give **15e**]; mp = 189 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.76 (m, 6 H), 1.06 (m, 1 H), 1.37 (m, 1 H), 1.64 (m, 1 H), 2.78−2.90 (m, 2 H), 3.13 (s, 3 H), 3.86 (t, 1 H, J = 8.4 Hz), 4.38 (t, 2 H, J = 6.0 Hz), 4.64 (dd, 1 H, J = 3.2, 10.8 Hz), 5.00 (ab quart, 2 H, J = 5.2, 13.6 Hz), 5.20 (ab quart, 2 H, J = 6.0, 12.4 Hz), 7.24−7.39 (m, 11 H), 8.53 (m, 1 H), 8.79 (t, 1 H, J = 6.0 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 10.92, 15.31, 24.35, 28.27, 30.52, 36.35, 36.49, 43.34, 58.91, 61.11, 62.89, 64.16, 65.39, 67.17, 126.41, 126.62, 127.69, 127.78, 127.97, 127.98, 128.04, 128.25, 128.34, 128.46, 135.21, 137.06, 142.53, 155.96, 165.29, 168.43, 171.42, 171.89; IR (Nujol mull) 3295, 1729, 1691, 1646, 1545 cm^{-1}; HRMSFAB$^+$ ($C_{27}H_{36}N_3O_8S$, $M^+ + 1$) Calcd: 562.2223 found: 562.2229.

Scheme A: General Procedure to Synthesize Final Zwitterions: A 5% formic acid in methanol solution was degassed with argon. Under a positive argon flow, the Pd-black catalyst (1:1 by weight) was slowly added to the vigorously stirring degassed solution. **15** was dissolved in a minimal amount of formic acid and was added to the suspension

of catalyst. After 15−25 min, the suspension was filtered through Celite; and, the Celite pad was washed with more of the formic acid solution, methanol, and with dIH_2O. The filtrate was concentrated under partial vacuum and the residue was crystallized from ethanol: ether. (In some cases a recrystallization from H_2O:ethanol was required to obtain a clean compound.) The resultant solid was collected by vacuum filtration and was dried in vacuo with heating over P_2O_5 to give the desired zwitterion**.**

***N*-[(3-*S*-Amino-2-phenylpropanoylamino)methyl] 2-methanesulfonyl succinamic acid (17):** 65 mg (0.11 mmol) of **15c**, 64 mg of Pd-black, 15 mL of 5% formic acid in methanol; yield 20 mg (50% from **15c**, 35% from **14c**) of a white solid; mp = 168 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 2.90 (m, 3 H), 3.20 (m and s, 5 H), 4.13 (t, 1 H, J = 7.6 Hz), 4.28 (m, 1 H), 4.52 (m, 2 H), 7.30 (d, 2 H, J = 6.8 Hz), 7.42 (m, 3 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 31.45, 37.23, 37.97, 43.64, 43.80, 53.59, 54.24, 67.55, 126.77, 128.37, 129.32, 129.44, 135.86, 136.20, 165.98, 169.87, 170.37, 170.92; IR (Nujol mull) br 3400−2500, 1691, 1621, 1576, 1532 cm^{-1}; HRMSFAB$^+$ ($C_{15}H_{22}N_3O_6S$, $M^+ + 1$) Calcd: 372.1229 found: 372.1231.

***N*-[(3-*S*-Aminomethylbutrylamino) methyl] 2-methanesulfonyl succinamic acid (18):** 114 mg (0.210 mmol) of **15d**, 125 mg of Pd-black, 10 mL of 5% formic acid in methanol (Note: due to the high insolubility of **15d** a significantly greater amount, 10 mL, of formic acid was required to dissolve **15d**); yield 45 mg (67% from **15d**, 36% from **14d**) of a white solid; mp = 176 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 1.03 (d, 6 H, J = 6.4 Hz), 2.20 (sextet, 1 H, J = 6.4 Hz), 2.95 (m, 2 H), 3.22 (s, 3 H), 3.75 (d, 1 H, J = 5.6 Hz), 4.32 (m, 1 H), 4.66 (m, 2 H); ^{13}C NMR (100 MHz, D_2O) δ 17.43, 18.04, 30.28, 32.43, 39.84, 44.73, 59.04, 68.03, 170.03, 172.50, 172.55; IR (Nujol mull) br 3300−2500, 1691, 1627, 1545, 1507, 1139 cm^{-1}; HRMSFAB$^+$ ($C_{11}H_{22}N_3O_6S$, $M^+ + 1$) Calcd: 324.1229 found: 324.1229.

***N*-[(3-*S*-Aminomethylpentanoylamino) methyl] 2-methanesulfonyl succinamic acid (19):** 90 mg (0.16 mmol) of **15e**, 94 mg of Pd-black, 15 mL of 5% formic acid in methanol; yield 36 mg (67% from **15e**, 62% from **14e**) of a white solid; mp = 155 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 0.949 (t, 3 H, J = 7.4 Hz), 1.00 (d, 3 H, J = 6.8 Hz), 1.24 (m, 1 H), 1.51 (m, 1 H), 1.95 (m, 1 H), 2.95 (m, 2 H), 3.22 (s, 3 H), 3.81 (dd, 1 H, J = 1.4, 6.8 Hz), 4.32 (m, 1 H), 4.65 (m, 2 H); ^{13}C NMR (100 MHz, D_2O) δ 10.96, 14.67, 24.60, 32.43, 32.48, 37.11, 39.84, 44.72, 58.39, 68.00, 170.05, 171.63, 172.44, 172.49; IR (Nujol mull) br 3300−2500, 1697, 1627, 1545, 1507, 1107 cm^{-1}; HRMSFAB$^+$ ($C_{12}H_{24}N_3O_6S$, $M^+ + 1$) Calcd: 338.1386 found: 338.1385.

Scheme B: General Procedure to Synthesize Final Zwitterion. In a dried flask under argon, 1 equiv of **14** was suspended in anhydrous dichloromethane. The suspension was chilled to approximately 0 °C. In a second dried flask under argon 2.2 equiv of *m*-chloroperbenzoic acid (mCPBA) was dissolved in anhydrous dichloromethane. The mCPBA solution was added in portions to the suspension of **14**. The reaction mixture was stirred for 3−4 h at 0 °C. When the oxidation was complete, 2−4 mL of a saturated sodium thiosulfate solution was added to the reaction mixture to quench the residual mCPBA. The reaction mixture was then stirred vigorously for 10−20 min at which time the reaction mixture was partitioned between dIH_2O and chloroform. The organics were washed three times with saturated aqueous $NaHCO_3$ solution and one time with brine. The organics were then dried over $MgSO_4$. After filtration the organics were concentrated in vacuo, and the resultant crude sulfone **15** was used directly in the hydrogenation reaction. A 5% formic acid in methanol solution was degassed with argon. Under a positive argon flow, the Pd-black catalyst (1:1 by weight) was slowly added to the vigorously stirring degassed solution. **15** was dissolved in a minimal amount of formic acid and was added to the suspension of catalyst. After 15−25 min the suspension was filtered through Celite, and the Celite pad was washed with more of the formic acid solution, methanol, and with dIH_2O. The filtrate was concentrated under partial vacuum, and the residue was crystallized from ethanol:ether. (In some cases a recrystallization from H_2O:ethanol was needed.) The resultant solid was collected by vacuum filtration and was dried in vacuo with heating over P_2O_5 to give the desired zwitterion**.**

***N*-[(2-Amino-acetylamino)methyl] 2-methanesulfonylsuccinamic acid (16):** 203 mg (0.420 mmol) of **14b**, 10 mL of dichloromethane, 271 mg (0.890 mmol) of mCPBA, 10 mL of dichloromethane, 115 mg of Pd-black, 15 mL of 5% formic acid in methanol; yield 42 mg (35% from **14b**) of a white solid; mp = 165−167 °C. ^{1}H NMR (400 MHz, D_2O) δ 2.94 (s, 1 H), 2.96 (d, 1 H, J = 3.2 Hz), 3.22 (s, 3 H), 3.81 (s, 2 H), 4.32 (t, 1 H, J = 7.0 Hz), 4.66 (ab quart, 2 H, J = 13.6, 4.0 Hz); ^{13}C NMR (100 MHz, D_2O) δ 32.54, 39.86, 40.98, 44.78, 68.07, 168.10, 170.08, 172.61; IR (Nujol mull) br 3350−2500, 1627, 1551 cm^{-1}; HRMSFAB$^+$ ($C_8H_{16}N_3O_6S$, $M^+ + 1$) Calcd: 282.0760 found: 282.0760.

2-Methanesulfonyl-*N*-{[(pyrrolidine-2-*S*-carbonyl)amino]methyl} succinamic acid (20): 125 mg (0.240 mmol) of **14f**, 5 mL of dichloromethane, 154 mg (0.530 mmol) of mCPBA, 5 mL of dichloromethane, 100 mg of Pd-black, 15 mL of 5% formic acid in methanol; yield 28 mg (36% from **14f**) of a white solid; mp = 173 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 2.05 (m, 3 H), 2.40 (m, 1 H), 2.94 (m, 2 H), 3.21 (s, 3 H), 3.40 (m, 2 H), 4.31 (m, 1 H), 4.64 (ab quart and s, 2 H, J = 13.6, 14.8 Hz); ^{13}C NMR (100 MHz, D_2O) δ 24.25, 29.99, 32.46, 32.53, 39.87, 44.93, 46.93, 60.23, 68.07, 170.04, 170.27, 170.30, 172.58; IR (Nujol mull) br 3350−2500, 1697, 1589, 1526, 1132 cm^{-1}; HRMSFAB$^+$ ($C_{11}H_{20}N_3O_6S$, $M^+ + 1$) Calcd: 322.1073 found: 322.1074.

***N*-[(2-*R*-Amino-propanoylamino)methyl] 2-methanesulfonyl succinamic acid (21):** 175 mg (0.360 mmol) of **14g**, 8 mL of dichloromethane, 260 mg (0.860 mmol) of mCPBA, 7 mL of dichloromethane, 110 mg of Pd-black, 10 mL of 5% formic acid in methanol; yield 51 mg (48% from **14g**) of a white solid; mp = 174 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 1.52 (d, 3 H, J = 6.8 Hz), 2.95 (m, 2 H), 3.22 (s, 3 H), 4.06 (quart, 1 H, J = 6.8 Hz), 4.32 (t, 1 H, J = 7.2 Hz), 4.64 (ab quart and s, 2 H, J = 13.6, 9.2 Hz); ^{13}C NMR (100 MHz, D_2O) δ 16.78, 32.52, 39.86, 44.86, 49.48, 68.06, 170.06, 171.39, 172.58; IR (Nujol mull) br 3400−2500, 1691, 1633, 1532, 1126 cm^{-1}; HRMSFAB$^+$ ($C_9H_{18}N_3O_6S$, $M^+ + 1$) Calcd: 296.0916 found: 296.0916.

***N*-[(3-*R*-Aminomethylpentanoylamino)methyl] 2-methanesulfonyl succinamic acid (22):** 85 mg (0.16 mmol) of **14h**, 8 mL of dichloromethane, 88 mg (0.34 mmol) of mCPBA, 9 mL of dichloromethane; yield 55 mg (61%) of a white solid [Note: a precipitate was observed in the organic layer; therefore, the organics were concentrated directly under partial vacuum. The resultant residue was triturated with ether to give the sulfone **15h**.]; 38 mg (0.070 mmol) of **15h**, 55 mg of Pd-black, 5 mL of 5% formic acid in methanol; yield 16 mg (70% from **15h**, 44% from **14h**) of a white solid; mp = 154 °C (dec). ^{1}H NMR (400 MHz, D_2O, partial data) δ 0.95 (t, 3 H, J = 7.4 Hz), 1.00 (d, 3 H, J = 7.2 Hz), 1.24 (m, 1 H), 1.51 (m, 1 H), 1.97 (m, 1 H), 2.95 (m, 2 H), 3.22 (s, 3 H), 3.84 (d, 1 H, J = 5.6 Hz), 4.31 (m, 1 H), 4.65 (m); ^{13}C NMR (100 MHz, D_2O) δ 10.40, 13.95, 24.06, 31.89, 36.09, 39.32, 44.22, 57.57, 67.50, 169.44, 172.00; IR (Nujol mull) br 3300−2500, br 1800−1600, br 1550−1500, 1296, 1142 cm^{-1}; HRMSFAB$^+$ ($C_{12}H_{24}N_3O_6S$, $M^+ + 1$) Calcd: 338.1386 found: 338.1385.

***N*-[1-*S*-(2-*S*-Amino-propanoylamino)ethyl] 2-methanesulfonyl succinamic acid (23):** 127 mg (0.250 mmol) of **14i**, 104 mg (0.550 mmol) of mCPBA, 10 mL of dichloromethane, 5 mL of 5% formic acid in methanol (NOTE: an unweighed portion of Pd-black was added directly to the degassed solution); yield 16 mg (21% from **14i**) of a white solid; mp = 165−167 °C. ^{1}H NMR (400 MHz, D_2O) δ 1.42 (d, 3 H, J = 6.4 Hz), 1.50 (m, 3 H), 2.92 (m, 2 H), 3.21 (s, 3 H), 4.00 (m, 1 H), 4.29 (m, 1 H), 5.63 (quintet, 1 H, J = 6.4 Hz); ^{13}C NMR (100 MHz, D_2O) δ 16.98, 19.43, 19.55, 32.54, 39.90, 49.58, 53.66, 68.13, 170.05, 171.25; IR (Nujol mull) br 3400−2500, br 1700−1520, 1284, 1119 cm^{-1}; HRMSFAB$^+$ ($C_{10}H_{19}N_3O_6S$, $M^+ + 1$) Calcd: 310.1073 found: 310.1073.

***N*-[1-*R*-(2-*S*-Amino-propanoylamino)ethyl] 2-methanesulfonyl succinamic acid (24):** 79 mg (0.16 mmol) of **14j**, 73 mg (0.40 mmol) of mCPBA, 8 mL of dichloromethane; 5 mL of 5% formic acid in methanol (NOTE: an unweighed portion of Pd-black was added directly to the degassed solution); yield 30 mg (61% from **14j**) of a white solid; mp = 189 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 1.41 (dd, 3 H, J = 3.2, 6.4 Hz), 1.51 (dd, 3 H, J = 4.8, 7.0 Hz), 2.93 (m, 2 H), 3.21 (s, 3 H), 4.00 (quart, 1 H, J = 7.0 Hz), 4.30 (t, 1 H, J = 6.4 Hz), 5.62 (m, 1 H); ^{13}C NMR (100 MHz, D_2O) δ 16.86, 19.43, 19.48, 32.50, 32.56, 39.89, 49.50, 53.70, 53.77, 68.03, 170.08, 171.48; IR (Nujol mull) br 3400−2500, br 1700−1520, 1297, 1132 cm^{-1}; HRMSFAB$^+$ ($C_{10}H_{19}N_3O_6S$, $M^+ + 1$) Calcd: 310.1073 found: 310.1073.

***N*-[2-(2-*S*-Amino-propanoylamino)ethyl] 2-Methanesulfonyl Succinamic Acid (25).** In 10 mL of anhydrous dichloromethane under

argon was suspended 80 mg (0.16 mmol) of **29**. The resultant suspension was chilled to 0−5 °C, and 65 mg (0.35 mmol) of mCPBA was added to the chilled suspension. The reaction mixture was stirred at 0−5 °C for 2 h. The reaction mixture was then diluted with dichloromethane. The organics were washed vigorously with saturated aqueous sodium thiosulfate. The organics were then washed three times with saturated aqueous $NaHCO_3$ and dried over $MgSO_4$. After filtering the organics were concentrated under partial vacuum. The resultant residue was dissolved in 2 mL of 5% formic acid in methanol. This solution was added to a suspension of Pd-black in 3 mL of degassed 5% formic acid in methanol. The reaction mixture was stirred vigorously for 15 min. The suspension was filtered through Celite to remove the catalyst, and the filtrate was concentrated on a rotary evaporator. After crystallization from methanol:ether at −25 °C, 17 mg (34%) of a whitish solid were collected by vacuum filtration and dried in vacuo; mp = 172−174 °C. ^{1}H NMR (400 MHz, D_2O) δ 1.35 (m, 3 H), 2.76 (m, 2 H), 3.05 (s, 3 H), 3.1−3.4 (m, 4 H), 3.87 (q, 1 H, J = 7.1 Hz), 4.14 (t, 1 H, J = 7.4 Hz); ^{13}C NMR (100 MHz, D_2O) δ 16.92, 32.81, 38.96, 39.42, 39.84, 49.63, 68.26, 170.15, 171.42, 172.41; IR ($CHCl_3$) 3300−2500 br, 1708−1500 br, 1294, 1132 cm^{-1}; HRMSFAB$^+$ ($C_{10}H_{18}N_3O_6S$, $M^+ + 1$) Calcd: 310.1073 found: 310.1073.

[1-*S*-(2-*tert*-Butoxycarbonylamino-ethylcarbamoyl)ethyl]carbamic Acid Benzyl Ester (27). In a flame-dried flask under an atmosphere of argon, 510 mg of *Z*-L-alanine (2.20 mmol) was dissolved in anhydrous tetrahydrofuran (20 mL). To the resultant solution was added 440 mg of 1,1′-carbonyldiimidazole (2.60 mmol) with stirring. After stirring 1−2 h at room temperature, 250 mg (1.60 mmol) of *tert*-butyl *N*-(2-aminoethyl) carbamate was added to the reaction mixture. The reaction mixture was stirred at room-temperature overnight. The tetrahydrofuran was then removed under partial vacuum. The resultant residue was partitioned between ethyl acetate and water. The organics were washed with saturated sodium bicarbonate and 1 N HCl. The organics were dried over $MgSO_4$. After filtration the organics were concentrated in vacuo, giving 350 mg (61%) of a white solid; mp = 161−163 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.18 (d, 3 H, J = 7.2 Hz), 1.37 (s, 9 H), 2.95−3.16 (m, 4 H), 3.97 (quintet, 1 H, J = 7.2 Hz), 5.01 (ab quart, 2 H, J = 8.8, 12.8 Hz), 6.75 (br s, 1 H), 7.35 (m, 6 H), 7.87 (br s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 18.20, 28.18, 30.38, 50.08, 65.32, 77.61, 127.70, 128.25, 136.97, 155.58, 172.44; IR (Nujol mull) 3349, 3305, 1690, 1649, 1538, 1169 cm^{-1}; HRMSFAB$^+$ ($C_{18}H_{28}N_3O_5$, $M^+ + 1$) Calcd: 366.2029 found: 366.2028.

[1-*S*-(2-Aminoethylcarbamoyl)ethyl]carbamic acid benzyl ester hydrochloride (28): 281 mg of **27** (0.770 mmol) was dissolved in 20 mL of methanol. To the resultant solution 1.0 mL of concentrated HCl was added. After stirring at room temperature for 22 h the reaction mixture was concentrated in vacuo; 224 mg (97%) of a white solid were isolated; mp = 173−174 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.22 (d, 3 H, J = 7.2 Hz), 2.83 (m, 2 H), 3.30 (m, 2 H), 4.00 (quintet, 1 H, J = 7.2 Hz), 5.02 (ab quart, 2 H, J = 10.4, 12.8 Hz), 7.35 (m, 5 H), 7.47 (d, 1 H, J = 7.2 Hz), 7.95 (br s, 3 H), 8.15 (br s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.99, 36.41, 38.41, 50.19, 65.42, 127.76, 128.32, 136.93, 155.70, 172.93; IR (Nujol mull) 3342, 3290, br 1690−1642, 1546, 1257 cm^{-1}; HRMSFAB$^+$ ($C_{13}H_{20}N_3O_3$, M^+ − Cl) Calcd: 266.1505 found: 266.1503.

***N*-[2-(2-*S*-Benzyloxycarbonylamino-propanoylamino)ethyl] 2-methanesulfanyl succinamic acid benzyl ester (29):** 192 mg (0.660 mmol) of **28** was suspended in 15 mL of anhydrous dichloromethane in a dried flask under argon. To the resultant suspension was added 130 μL (1.00 mmol) of *N*-ethylmorpholine. The reaction mixture was stirred for 5 min before a solution of **13a** (162 mg, 0.660 mmol) in 5 mL of anhydrous dichloromethane was added. Finally, 134 mg (0.860 mmol) of HOBT and 150 mg (0.800 mmol) of EDAC were added to the reaction mixture. After 24 h the reaction mixture was diluted with ethyl acetate, and the organics were washed with 1 N HCl and saturated sodium bicarbonate solution. The organics were dried over $MgSO_4$. After the reaction mixture was filtered, the organics were concentrated in vacuo, giving 247 mg (77%) of a beige solid; mp = 104−107 °C. ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.18 (d, 3 H, J = 7.2 Hz), 2.07 (s, 3 H), 2.73 (dd, 1 H, J = 9.2, 16.0 Hz), 3.00−3.14 (m, 4 H), 3.64 (dd, 1 H, J = 6.0, 9.2 Hz), 3.96 (quintet, 1 H, J = 7.2 Hz), 5.0−5.2 (m, 4 H), 7.36 (m, 6 H), 7.87 (br s, 1 H), 7.95 (br s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.43, 18.14, 36.49, 38.21, 42.33, 50.13, 65.34, 65.86, 127.69, 127.95, 128.07, 128.28, 128.35, 136.01, 136.97, 155.63, 169.13, 170.94, 172.49; IR ($CHCl_3$) 3308, 3086, 3000−2940 br, 1722, 1659, 1532, 1450, 1240 cm^{-1}; HRMSFAB$^+$ ($C_{25}H_{32}N_3O_6S$, $M^+ + 1$) Calcd: 502.2012 found: 502.2012.

***N*-[(2-*S*-Amino-propanoylamino)methyl] 2-*S*-Methanesulfanyl Succinamic Acid (30).** To a degassed 5% formic acid in methanol solution (3 mL), was added 50 mg of Pd-black. To the vigorously stirred suspension a solution of **14k**, 56 mg (0.11 mmol) in 2 mL of 5% formic acid in methanol, was added. After 30 min the suspension was filtered through Celite to remove the palladium catalyst. The filtrate was concentrated under partial vacuum. After crystallization from methanol: ether, 10 mg (35%) of a white solid were isolated; mp = 148−152 °C (dec). ^{1}H NMR (400 MHz, D_2O, partial data) δ 1.53 (d, 3 H, J = 7.0 Hz), 2.13 (s, 3 H), 2.59 (dd, 1 H, J = 8.0, 14.8 Hz), 2.79 (dd, 1 H, J = 7.0, 14.8 Hz), 3.55 (t, 1 H, J = 8.0 Hz), 4.06 (q, 1 H, J = 7.0 Hz), 4.6 (m); ^{13}C NMR (100 MHz, D_2O) δ 13.60, 16.82, 38.49, 44.78, 47.49, 49.49, 171.30, 174.35; IR (Nujol mull) br 3200−2500, 1669, br 1600−1500, 1210, 1117 cm^{-1}; HRMSFAB$^+$ ($C_9H_{17}N_3O_4S$, $M^+ + 1$) Calcd: 264.1018 found: 264.1017.

***N*-[(2-*S*-Amino-propanoylamino)methyl] 2-*R*-Methanesulfanyl Succinamic Acid (31).** To a degassed 5% formic acid in methanol solution (3 mL) was added 62 mg of Pd-black. To the vigorously stirred suspension a solution of **14a**, 55 mg (0.11 mmol) in 2 mL of 5% formic acid in methanol, was added. After 30 min the suspension was filtered through Celite to remove the palladium catalyst. The filtrate was concentrated under partial vacuum. After crystallization from methanol: ether, 18 mg (63%) of a white solid were isolated; mp = 183−185 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 1.51 (d, 3 H, J = 7.2 Hz), 2.57 (dd, 1 H, J = 7.8, 15.2), 2.75 (dd, 1 H, J = 7.8, 15.2 Hz), 3.53 (t, 1 H, J = 7.8 Hz), 4.04 (q, 1 H, J = 7.2 Hz), 4.60 (m, 2 H); ^{13}C NMR (100 MHz, D_2O) δ 13.60, 16.89, 38.56, 44.76, 47.77, 49.51, 171.50, 174.48; IR (Nujol mull) 3281, br 3200−2500, 1686, 1555, 1248, 1099 cm^{-1}; HRMSFAB$^+$ ($C_9H_{17}N_3O_4S$, $M^+ + 1$) Calcd: 264.1018 found: 264.1017.

***N*-[(2-*S*-Amino-propanoylamino)methyl] 2-Methanesulfinyl Succinamic Acid (32).** In a dried flask under argon 69 mg (0.14 mmol) of **14a** was dissolved in 5 mL of anhydrous dichloromethane. To the resultant mixture 26 mg (0.16 mmol) of mCPBA was added, and the reaction mixture was stirred for 30 min at room temperature. A saturated sodium thiosulfate solution (∼5 mLs) was added to quench residual mCPBA. The organics were then washed with an aqueous saturated $NaHCO_3$ solution and then were dried over $MgSO_4$. After filtration, the organics were concentrated in vacuo. The resultant crude sulfoxide was dissolved in 2 mL of a 5% formic acid in methanol solution. The resultant solution was added to a vigorously stirred suspension of Pd-black in 3 mL of degassed 5% formic acid in methanol solution. After approximately 30 min the suspension was filtered through Celite to remove the palladium catalyst. The filtrate was concentrated under partial vacuum. After crystallization from methanol:ether, 22 mg (56%) of a white solid was isolated; mp = 163−165 °C. ^{1}H NMR (400 MHz, D_2O, partial data) δ 1.52 (d, 3 H, J = 7.4 Hz), 2.6−3.0 (2s and m, 5 H), 3.84 (m, 1 H), 4.06 (q, 1 H, J = 7.4 Hz), 4.16 (dd, 1 H, J = 4.6, 5.2 Hz), 4.64−4.70 (m); ^{13}C NMR (100 MHz, D_2O) δ 16.82, 30.57, 33.00, 33.54, 37.30, 44.88, 44.94, 49.51, 63.60, 66.97, 171.39, 173.17, 173.63; IR (Nujol mull) br 3200−2500, br 1670−1540, 1236, 1108, 1012 cm^{-1}; HRMSFAB$^+$ ($C_9H_{18}N_3O_5S$, $M^+ + 1$) Calcd: 280.0967 found: 208.0966.

Fmoc-L-alanylglycinamide (34). In a dried flask under argon 314 mg (1.00 mmol) of glycinamide hydrochloride was suspended in 10 mL of anhydrous dichloromethane. To the resultant suspension was added 170 μL (1.00 mmol) of anhydrous diisopropylethylamine, and the reaction mixture was stirred for 20 min at room temperature. To the resultant suspension were then added 236 mg (1.20 mmol) of EDAC and 147 mg (1.10 mmol) of HOBT. Finally 314 mg (1.00 mmol) of Fmoc-L-alanine were added to the reaction mixture. The reaction mixture was stirred overnight under argon, resulting in a viscous suspension. The dichloromethane was removed on the rotary evaporator, and the resultant residue was triturated with an aqueous 4% $NaHCO_3$ solution. A fine white solid was collected by vacuum filtration and dried in vacuo, yielding 160 mg (43%) of **34**; mp = 143−145 °C. ^{1}H

NMR (400 MHz, DMSO-d_6) δ 1.22 (d, 2 H, J = 7.2 Hz), 3.62 (t, 2 H, J = 6.0 Hz), 4.04 (quintet, 1 H, J = 7.2 Hz), 4.26 (m, 3 H), 7.06 (br s, 1 H), 7.17 (br s, 1 H), 7.33 (t, 2 H, J = 7.4 Hz), 7.41 (*t*, 2 H, J = 7.4 Hz), 7.61 (d, 1 H, J = 7.2 Hz), 7.72 (t, 2 H, J = 6.6 Hz), 7.89 (d, 2 H, J = 7.6 Hz), 8.07 (br s, 1 H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 17.84, 41.92, 46.60, 50.19, 65.66, 109.73, 119.99, 120.17, 121.34, 125.25, 127.04, 127.26, 127.60, 128.89, 137.39, 139.38, 140.67, 143.56, 143.73, 155.86, 170.80, 172.59; IR (Nujol mull) 3432, 3300, 3213, 1693, 1658, 1544 cm^{-1}; HRMSFAB$^+$ ($C_{20}H_{21}N_3O_4$, $M^+ + 1$) Calcd: 368.1610 found: 368.1610.

[1-*S*-(Aminomethyl-carbamoyl)-ethyl]carbamic Acid 9-*H*-Fluoren-9-ylmethyl Ester Hydrochloride (35). The synthesis of the Fmoc derivative was conducted using the general procedure to synthesize geminal amino amides: 120 mg (0.330 mmol) of **34**, 154 mg (0.350 mmol) of PIFA, 2 mL of acetonitrile, 2 mL of H_2O. (Note: The aqueous layer was concentrated in vacuo, and the material was used directly without further purification.) Yield 109 mg (88%) of a beige solid; mp = 131 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6, partial data) δ 1.25 (d, 3 H, J = 7.2 Hz), 4.0−4.2 (m, 6 H), 7.33 (t, 2 H, J = 7.4 Hz), 7.42 (t, 2 H, J = 7.4 Hz), 7.67 (d, 1 H, J = 7.2 Hz), 7.72 (m, 2 H), 7.89 (d, 1 H, J = 7.6 Hz), 8.15 (br s, 3 H), 8.87 (br s, 1 H); ^{13}C NMR(100 MHz, DMSO-d_6) δ 17.75, 44.66, 46.59, 49.86, 65.64. 120.11, 125.24, 127.04, 127.63, 140.69, 143.73, 143.83, 155.72, 174.06; IR (Nujol mull) 3500−2500 br, 1681, 1532 cm^{-1}; HRMSFAB$^+$ ($C_{19}H_{22}N_3O_3$, M^+ − Cl) Calcd: 340.1661 found: 340.1660.

(*Z*)-3-(2-[2-S-9-*H*-Fluoren-9-ylmethoxycarbonylamino-propanoylamino]methyl) Acrylic Acid (37). In a dried flask under argon, 39 mg (0.10 mmol) of **35** were suspended in 5 mL of anhydrous dichloromethane. To the resultant suspension was added 12 mg (0.12 mmol) of maleic anhydride. Finally, 20 μL (0.10 mmol) of diisopropylethylamine was added to the suspension, and a clear solution resulted. The reaction mixture was stirred for 30 min at room temperature. The reaction mixture was diluted with ethyl acetate and was washed with 1 N HCl. The organics were dried over $MgSO_4$ and were concentrated in vacuo to give 35 mg (78%) of a white solid; mp = 171 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.20 (d, 3 H, J = 7.2 Hz), 4.03 (quintet, 1 H, J = 7.2 Hz), 4.24 (m, 3 H), 4.94 (m, 2 H), 6.30 (ab quart, 2 H, J = 12.6, 15.6 Hz), 7.33 (t, 2 H, J = 7.4 Hz), 7.41 (t, 2 H, J = 7.4 Hz), 7.50 (d, 1 H, J = 7.2 Hz), 7.72 (t, 2 H, J = 5.8 Hz), 7.89 (d, 2 H, J = 7.2 Hz), 8.54 (br s, 1 H), 9.35 (br s, 1 H); ^{13}C NMR(100 MHz, DMSO-d_6) δ 18.08, 43.65, 46.62, 49.83, 65.57, 120.07, 125.26, 127.04, 127.60, 130.18, 132.91, 140.68, 143.77, 155.61, 165.15, 166.05, 173.20; IR (Nujol mull) 3370−3300 br, 1714, 1664, 1631, 1590 cm^{-1}; HRMSFAB$^+$ ($C_{23}H_{24}N_3O_6$, $M^+ + 1$) Calcd: 438.1665 found: 438.1664.

(*Z*)-3-(*N*-[(2-*S*-Aminopropanoylamino)methyl])acrylic acid (38): 40 mg (0.090 mmol) of **37** was dissolved in 1 mL of dimethylformamide. To the resultant solution was added 100 μL of diethylamine. After 1.5 h at room temperature the reaction mixture was diluted with dIH_2O and was washed exhaustively with ethyl acetate. The aqueous extract was concentrated under partial vacuum. After crystallization from methanol:ether, a white solid was collected by vacuum filtration and was dried in vacuo to give 12 mg (62%): mp = 173 °C (dec). ^{1}H NMR (400 MHz, D_2O) δ 1.53 (d, 3 H, J = 6.8 Hz), 4.06 (q, 1 H, J = 6.8 Hz), 4.69 (s, 2 H), 5.95 (d, 1 H, J = 12.6 Hz), 6.39. (d, 1 H, J = 12.6 Hz); ^{13}C NMR(100 MHz, D_2O) δ 16.79, 44.75, 49.50, 124.07, 137.54, 168.75, 171.49, 175.00; IR (Nujol mull) 3500−2500 br, 1672, 1600−1500 br cm^{-1}; HRMSFAB$^+$ ($C_8H_{14}N_3O_4$, $M^+ + 1$) Calcd: 216.0984 found: 216.0985.

(*E*)-3-{2-[2-S-9-*H*-Fluoren-9-ylmethoxycarbonylamino-propanoylamino]methyl}acrylic Acid Methyl Ester (40). In a dried flask under argon, **35** (80 mg, 0.21 mmol) was suspended in 10 mL of anhydrous dichloromethane. To the resultant suspension was added 25 mg (0.19 mmol) of methyl hydrogen fumerate. To the resultant reaction mixture was added 40 μL (0.20 mmol) of anhydrous diisopropylethylamine. Finally, 52 mg (0.24 mmol) of EDAC and 37 mg (0.26 mmol) of HOBT were added to the reaction mixture. After 20 h total reaction time, the reaction mixture was diluted with ethyl acetate. The organics were washed with 1 N HCl and saturated aqueous $NaHCO_3$. The organics were concentrated on the rotary evaporator, and the resultant residue was triturated with methanol and dIH_2O. The resultant cream solid was collected by vacuum filtration and dried in vacuo giving 60 mg (70%); mp = 204 °C (dec). ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.19 (d, 3 H, J = 6.8 Hz), 3.71 (s, 3 H), 4.03 (t, 1 H, J = 6.8 Hz), 4.24 (m, 3 H), 4.50 (q, 2 H, J = 6.0 Hz), 6.60 (d, 1 H, J = 15.2 Hz), 7.02 (d, 1 H, J = 15.2 Hz), 7.32 (t, 2 H, J = 7.0 Hz), 7.41 (t, 2 H, J = 7.6 Hz), 7.49 (d, 1 H, J = 7.2 Hz), 7.71 (t, 2 H, J = 6.8 Hz), 7.88 (d, 2 H, J = 7.2 Hz), 8.53 (br s, 1 H), 9.11 (br s, 1 H); ^{13}C NMR(100 MHz, DMSO-d_6) δ 18.08, 43.58, 46.62, 49.85, 52.00, 65.56, 120.07, 125.25, 127.04, 127.59, 128.70, 137.21, 140.68, 143.77, 155.61, 163.06, 165.37, 173.13 IR (Nujol mull) 3298, 3290, 3067, 1720, 1689, 1648, 1557 cm^{-1}; HRMSFAB$^+$ ($C_{24}H_{27}N_3O_6$, $M^+ + 1$) Calcd: 452.1822 found: 452.1822.

(*E*)-3-(*N*-[(2-*S*-Aminopropanoylamino)methyl])acrylic acid (41): 42 mg (0.090 mmol) of **40** was suspended in 2 mL of tetrahydrofuran, and the reaction mixture was chilled 0−5 °C. To the chilled suspension was added 1.7 mL of 0.1 M LiOH solution dropwise. After 30 min total reaction time, the resultant solution was acidified to pH = 2 with 1 N HCl. The acidified reaction mixture was extracted with ethyl acetate. The organics were washed with brine and were then concentrated under partial vacuum. The resultant residue was suspended in 2 mL of dimethylformamide. To the resultant suspension was added 80 μL diethylamine with stirring. After an hour, the reaction mixture was diluted with dIH_2O and was washed exhaustively with ethyl acetate. The aqueous extract was concentrated under partial vacuum. After two successive crystallizations from methanol:ether, 9 mg (46%) of a white solid was isolated; mp = 210 °C (dec). ^{1}H NMR (400 MHz, D_2O, partial data) δ 1.50 (d, 3 H, J = 6.8 Hz), 4.02 (q, 1 H, J = 6.8 Hz), 6.63 (d, 1 H, J = 15.6 Hz), 6.78 (d, 1 H, J = 15.6 Hz); ^{13}C NMR(100 MHz, D_2O) δ 17.14, 44.99, 49.60, 130.93, 138.02, 168.53, 172.17, 173.83; IR (Nujol mull) 3290−2500 br, 1691, 1640, 1532 cm^{-1}; HRMSES$^+$ ($C_8H_{14}N_3O_4$, $M^+ + 1$) Calcd: 216.0984 found: 216.0998.

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Supporting Information Available: ^{13}C NMR spectra of compounds **16**−**22**, **23**−**25**, **30**−**32**, **38** and **41** (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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