Solid-Phase Synthesis of α,α-Difluoro-β-amino Acids via the Reformatsky Reaction

Agnès Vidal, Adel Nefzi, and Richard A. Houghten*

Torrey Pines Institute for Molecular Studies, San Diego, California 92121

rhoughten@tpims.org

Received August 27, 2001

Introduction

The rapid synthesis of large libraries of low molecular weight compounds by combinatorial methods has attracted enormous attention as a means of accelerating the drug discovery process.[1] Among potential candidates, fluorinated compounds have been of great interest to medicinal chemists due to the unique physical and biological properties imparted by fluorine.[2] As a result, a large number of therapeutic agents containing strategically placed fluorine atoms are currently widely used.[3] The role of amino acids in biological functions is well established, and the area of fluorine-containing amino acids is rapidly expanding, taking an important place in the family of unusual amino acids.[4] α-Amino acids containing fluorine in the β-position have been shown to exhibit a wide range of biological effects.[5] More recently, the synthesis of β-fluoroalkyl β-amino acids, with the fluorine located in the γ-position relative to carboxyl functionality, has been reported.[6] Given the biological influence of fluorine substitution in the β-position relative to the amine and the biomedicinal and synthetic potential reported for β-amino acids,[7] we became interested in the synthesis of α,α-difluoro-β-amino acids.

Several methods have been described for the selective and efficient incorporation of fluorine into organic compounds in solution. Direct introduction of fluorine can be achieved with special fluorinating agents (DAST, SF_4, CF_3OF, HF, and others).[8] However, their use is limited to specific compounds because of their high reactivity. In addition, many of these reagents are expensive, toxic, and hazardous. Consequently, with the increasing accessibility of fluorinated building blocks, the CF_2-synthon approach has blossomed.

Herein, we report the first solid-phase parallel synthesis of fluorinated compounds. Our approach is based on the Reformatsky reaction using ethyl bromodifluoroacetate. First reported in 1984[9] and since widely applied in the synthesis of fluorinated bioactive compounds,[10] the Reformatsky reaction is now commonly used for the incorporation of the $-CF_2CO-$ group. Ethyl bromodifluoroacetate is employed to generate the α,α-difluoro Reformatsky reagent $BrZnCF_2CO_2Et$.[11] This intermediate reacts with aldehydes and ketones to provide 2,2-difluoro-3-hydroxy esters, hence providing a means for two-carbon homologation with a difluoro moiety under mild conditions. Using imines as substrates, lactams are usually formed in good yield.[12]

Starting from a polymer-supported amino acid, we describe here the solid-phase synthesis of α,α-difluoro-β-amino acids via the formation of the benzotriazolyl derivative using an aldehyde and subsequent Reformatsky reaction using ethyl bromodifluoroacetate.

Results and Discussion

The syntheses of all the described compounds were carried out on the solid-phase using the "tea-bag" method.[1b] Starting material **1** was prepared by coupling Boc-protected amino acids to *p*-methylbenzhydrylamine (MBHA) resin using standard peptide chemistry and subsequent cleavage of the Boc-protecting group in the presence of TFA.

Classical conditions for imine formation on the solid-phase involve the condensation of an amine and an aldehyde in the presence of a drying reagent such as trimethylorthoformate.[13] Treatment of an MBHA-linked amino acid **1** (Scheme 1) with an aldehyde, followed by the reaction with $BrZnCF_2CO_2Et$ generated in situ by mixing Zn and $BrCF_2CO_2Et$ in refluxing tetrahydrofuran, afforded, after saponification and HF cleavage, the cor-

(1) (a) Geysen, H. M.; Meloen, R. H.; Barteling, S. *J. Proc. Natl. Acad. Sci. U.S.A.* **1984**, *81*, 3998. (b) Houghten, R. A. *Proc. Natl. Acad. Sci. U.S.A.* **1985**, *82*, 5131. (c) Houghten, R. A.; Pinilla, C.; Blondelle, S. E.; Appel, J. R.; Dooley, C. T.; Cuervo, J. H. *Nature* **1991**, *345*, 8486. For recent reviews on molecular diversity and combinatorial libraries, see: (d) Weber, L. High-diversity combinatorial libraries. *Curr. Opin. Chem. Biol.* **2000**, *4*, 295. (e) Dolle, R. E.; Nelson, K. H., Jr. *J. Comb. Chem.* **1999**, *1*, 235. (f) Dolle, R. E. *Mol. Diversity* **1998**, *3*, 199.

(2) For a general discussion of biological activity and the importance of fluorinated amino compounds, see: Welch, J. T.; Eswarakrischnan, S. *Fluorine in Bioorganic Chemistry*; Wiley: New York, 1991.

(3) (a) Dollery, C. *Therapeutic Drugs*; Churchill Livingstone: Edinburgh, U.K., 1999.

(4) (a) *Fluorine-Containing Amino Acids*: *Synthesis and Properties*; Kukhar, V. P., Soloshonok, V. A., Eds.; Wiley: Chichester, 1995. (b) Soloshonok, V. A.; Soloshonok, I. V.; Kukhar, V. P.; Svedas, V. K. *J. Org. Chem.* **1998**, *63*, 1878. (c) Angelastro, M. R.; Bey, P.; Mehdi, S.; Peet, N. P. *Bioorg. Med. Chem. Lett.* **1992**, *2*, 1235. (d) Wanner, M. J.; Hageman, J. M.; Koomen, G.-J.; Pandit, U.K. *J. Med. Chem.* **1980**, *23*, 85. (e) Thaisrivongs, S.; Schostarez, H. J.; Pals, D. T.; Turner, S. R. *J. Med. Chem.* **1987**, *30*, 1837.

(5) Sewald N.; Burger K. *Fluorine-Containing Amino Acids*: *Synthesis and Properties*; Kukhar, V. P., Soloshonok, V. A., Eds.; Wiley: Chichester, 1995; pp 139−220.

(6) Soloshonok, V. A.; Soloshonok, I. V. *J. Org. Chem.* **1998**, *63*, 1878.

(7) (a) Appella, D. H.; Christianson, L. A.; Klein, D. A.; Powell, D. R.; Huang, X.; Barchi, J. J., Jr.; Gellman S. H. *Nature* **1997**, *387* (22 May), 381. (b) Borgman, S. *Chem. Eng. News* **1997**, June 16, 32.

(8) Wilkinson, J. A. *Chem. Rev.* **1992**, *92*, 505.

(9) Hallinan, E. A.; Fried, J. *Tetrahedron Lett.* **1984**, *25*, 2301.

(10) (a) Welch, J. T. *Tetrahedron* **1987**, *43*, 3123. (b) Doherty, A. M.; Sicar, I.; Kornberg, B. E.; Quin, J.; Winters, R. T.; Kaltenbronn, J. S.; Taylor, M. D.; Batley, B. L.; Rapundalo, S. R.; Ryan, M. J.; Painchaud, C. A. *J. Med. Chem.* **1992**, *35*, 2. (c) Schirlin, D.; Baltzer, S.; Altenburger, J. M. *Tetrahedron Lett.* **1988**, *29*, 3687. (d) Schirlin, D.; Baltzer, S.; Altenburger, J. M. *Tetrahedron Lett.* **1991**, *32*, 7255. (e) Robinson, R. P.; Donahue, K. *J. Org. Chem.* **1992**, *57*, 7309. (f) Thaisrivongs, S.; Pals, D. T.; Kati, W. M.; Turner, S. R.; Thomasco, L. M.; Watt, W. *J. Med. Chem.* **1986**, *29*, 2080. Thaisrivongs, S.; Pals, D. T.; Kati, W. M.; Turner, S. R.; Thomasco, L. M. *J. Med. Chem.* **1985**, *28*, 1555. (g) Peet, N. P.; Burkhart, J. P.; Angelastro, M. R.; Giroux, E.; L., Mehdi, S.; Bey, P.; Kolb, M.; Neises, B.; Schirlin, D. *J. Med. Chem.* **1990**, *33*, 394. (h) Hertel, L. W.; Kroin, J. S.; Misner, J. W.; Tustin, J. M. *J.Org. Chem.* **1988**, *53*, 2406. (i) Witkowski, S.; Rao, Y. K.; Premchandran, R. H.; Halushka, P. V.; Fried, J. *J. Am. Chem. Soc.* **1992**, *114*, 8464.

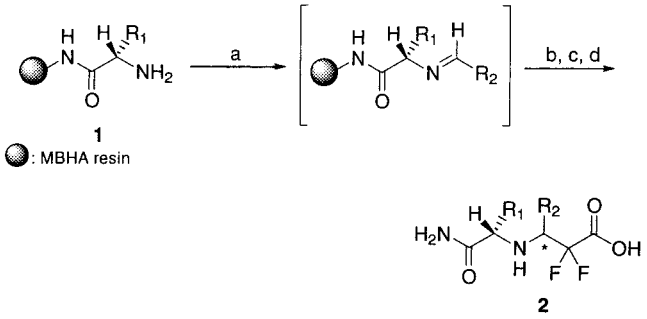
(11) (a) Burton, D. J.; Yang, Z,-Y. In *Chemistry of Organic Fluorine Compounds II: A Critical Review*; Hudlický, M., Pavlath, A. E., Eds.; ACS Monograph 187; American Chemical Society: Washington, DC, 1995; p 684. (b) Shen, Y.; Qi, M. *J. Fluorine Chem.* **1994**, *67*, 229.

(12) Taguchi, T.; Kitagawa, O.; Suda, Y.; Ohkawa, S.; Hashimoto, A.; Iitaka, Y.; Kobayashi, Y. *Tetrahedron Lett.* **1988**, *29*, 5291.

(13) Look, G. C.; Murphy, M. M.; Campbell, D. A.; Gallop, M. A. *Tetrahedron Lett.* **1995**, *36*, 2937.

10.1021/jo010872z CCC: $20.00
Published on Web 11/03/2001

Scheme 1[a]



[a] Key: (a) R_2CHO, trimethylorthoformate, DCM, rt; (b) Zn, $BrCF_2CO_2Et$, THF, reflux; (c) $LiOH·H_2O$; (iv) HF/anisole.

responding α,α-difluoro-*β*-amino acid **2** in low yields with various amounts of starting amine and other byproducts.[14]

As we were looking for a more efficient way to prepare and use imine derivatives, we decided to investigate the work of Katritzky et al.,[15] who successfully used the Reformatsky reaction of bromofluoroacetates with *N*-(α-aminoalkyl)benzotriazole for the preparation of α-fluoro-*β*-amino esters in solution. Another application of these Reformatsky reagents was their successful conversion to difluoroketene silyl acetals prepared in situ by the reaction of the zinc reagent with trimethylsilyl chloride.[12,16]

Following Katritzky's approach, we prepared the *N*-(α-aminoalkyl)benzotriazoles **3** (Scheme 2) by treating the resin-bound amino acids **1** with various aldehydes and benzotriazole, in refluxing benzene with a Dean−Stark trap, which were then used as iminium salt precursors[17] to obtain the α,α-difluorinated *β*-amino esters **4** via a Reformatsky reaction with ethyl bromofluoroacetate (14 equiv), zinc, and trimethylsilyl chloride in refluxing tetrahydrofuran. Following saponification of the ethyl ester in the presence of a lithium hydroxide solution and cleavage from the resin with HF, the corresponding α,α-difluorinated *β*-amino acids were obtained as a mixture of diastereoisomers **2** and **2′**. We examined the general applicability of the reaction using model compounds derived from five L-amino acids (Phe, Val, Ala, Gly, and Leu) and six aldehydes (benzaldehyde, 2-nitrobenzaldehyde, 2-fluorobenzaldehyde, *p*-methoxybenzaldehyde, 2,5-dimethylbenzaldehyde, and 3-furaldehyde).[18] We report here the results obtained for seven selected compounds (Table 1), which have been purified and fully characterized by LC-MS (ESI) and NMR (^1H, ^{13}C, COSY, HMBC, HMQC, and ^{19}F). For each aldehyde, two diastereoisomers were obtained, except for the glycine derivative (Table 1, entry 4) that gave nonseparable enantiomers. The relative configurations of the diastereoisomers were determined by NOE difference spectroscopy. Diastereoisomeric ratios were dependent on the nature of the aldehyde. The best selectivity was obtained using benzaldehyde with a diastereoisomeric excess greater than 90%. However, using a 14 equiv excess of Reformatsky reagent, two additional products **6** and **7** were formed (Scheme 3). Following cleavage from the solid support, the characterization of the resulting compounds by LC-MS showed that they had molecular weights +96 Da and +52 Da, respectively, greater than the desired compound **2**.

Reflux of the resin-bound intermediates **5**−**7** in ethanol, followed by cleavage from the solid support, led only to the desired product **2** and byproduct **8**, which had a molecular weight of 52 Da greater than expected. Further characterization of **8g** (R_1 = $PhCH_2$−, R_2 = Ph−) by ^1H, ^{13}C, and ^{19}F NMR showed that **8** resulted from the overaddition of the Reformatsky reagent on the newly formed ester **4**, followed by the decarboxylation of the intermediary compound **6**. This reaction, resulting from the enhanced reactivity of the ester carbonyl group, has been observed and reported with lithium enolates prepared from *tert*-butyl difluoroacetate.[19] The high electronegativity of fluorine atoms increases the electrophilicity of the carbonyl in the α-position and a consequent propensity for the formation of stable hydrates and hemiacetals.[20] To avoid or limit this double reaction, several experiments were performed using 2 and 6 equiv of $BrCF_2CO_2Et$. It appeared from LC-MS of cleaved controls that neither **6** or **7** were formed. However, the Reformasky coupling was not complete. As a result, imidazolidinone products were obtained following HF cleavage of the unreacted *N*-(α-aminoalkyl)benzotriazoles **3** by the nucleophilic substitution of the benzotriazole group with the amidic nitrogen. Further experiments were conducted to determine the optimal conditions for the formation of α,α-difluoro-*β*-amino acids (i.e., without the cyclization of the Bt derivatives to imidazolidinones or formation of tetrafluorinated compounds). Using 2, 6, 8, 10, and 12 equiv of the Reformatsky reagent, resin-bound amino acids (Phe, Val, Ala, Gly, and Leu), and 20 equiv of benzotriazole and benzaldehyde, we found that the use of 10 equiv of the Reformatsky reagents allowed the complete reaction of the benzotriazolyl derivatives without showing any formation of tetrafluorinated or cyclic compounds (Table 2). In an attempt to perform the Reformatsky reaction on resin-bound secondary amino acids **9** (proline or N-alkylated amino acids), we observed the complete formation of N-alkylated imidazolidinones **11**. No Reformatsky product **10** was detected (Scheme 4, Table 3). Further studies to determine whether the cyclization occurs before or after reaction with benzotriazole are under investigation. The solid-phase synthesis of a trisubstituted imidazolidinone library is the subject of a separate paper.[21]

Conclusion

We have explored a solid-phase method for the parallel conversion of a large number of primary amines to α,α-difluoro-*β*-amino acids by condensation of amines, aldehydes, benzotriazole and a Reformatsky reagent prepared

(14) The mass spectrum indicates the presence of the product of nucleophilic addition of the unreacted primary amine to the intermediary imine.

(15) Katritzky, A. R.; Nichols, D. A.; Qi, M. *Tetrahedron Lett.* **1998**, *39*, 7063.

(16) (a) Kitagawa, O.; Hashimoto, A.; Kobayashi, Y.; Taguchi, T. *Chem. Lett.*, **1990**, 1307. (b) Kitagawa, O.; Taguchi, T.; Kobayashi. Y. *Tetrahedron Lett.* **1988**, *29*, 1803.

(17) Katritzky, A. R.; Lan, X.; Yang, J. Z.; Denisko, O. V. *Chem. Rev.* **1998**, *98*, 409.

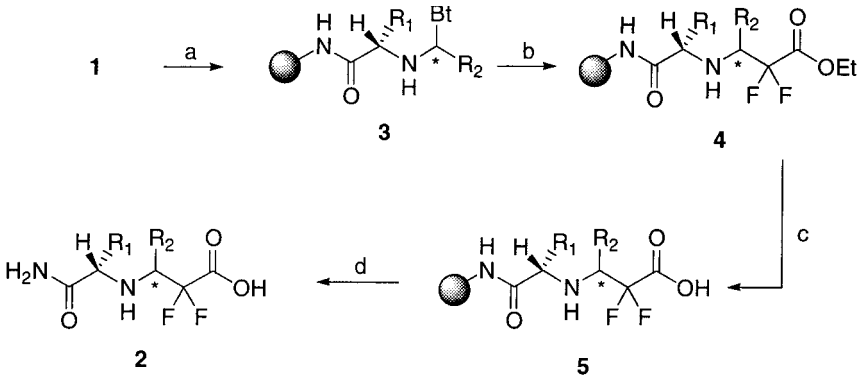
(18) Aromatic derivatives were selected for easy UV monitoring of the reaction.

(19) Taguchi, T.; Kitagawa, O.; Morikawa, T.; Nishiwaki, T.; Uehara, H.; Endo, H.; Kobayashi, Y. *Tetrahedron Lett.* **1986**, *27*, 6103.

(20) Bégué, J. P.; Bonnet-Delpon, D. *Tetrahedron* **1991**, *47*, 3207.

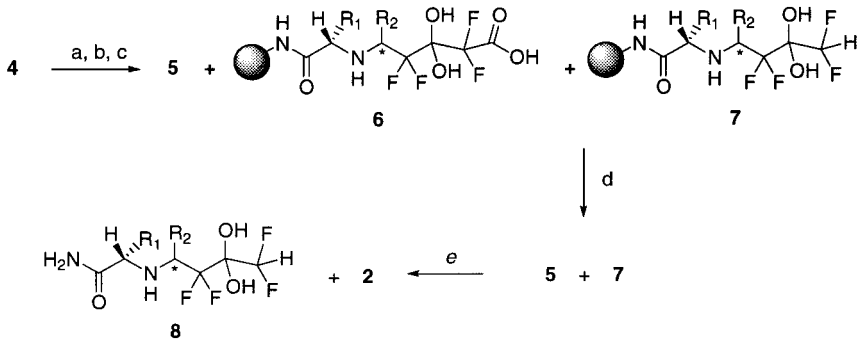
(21) Rinnová, M.; Vidal, A.; Nefzi, A.; Houghten, R. A. Solid-phase synthesis of 1,2,5-trisubstituted 4-imidazolidinones. Manuscript in publication.

Scheme 2[a]



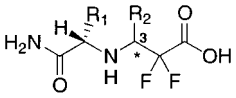
[a] Key: (a) BtH, R_2CHO, benzene, reflux; (b) Zn, TMSCl, $BrCF_2CO_2Et$, THF, reflux; (c) $LiOH.H_2O$, THF, rt; (d) HF/anisole.

Scheme 3[a]



[a] Key: (a) BtH, R_2CHO, benzene, reflux; (b) $BrZnCF_2CO_2Et$, TMSCl, THF, reflux; (c) $LiOH{\cdot}H_2O$, THF, rt; (d) EtOH, reflux; (e) HF/anisole.

Table 1. Examples of Compounds 2 Synthesized on a Solid Support



entry	compds	R_1	R_2	t_R[a] (**2x**/**2x′**) (ratio)[c]: configuration[b]	mass (M + H^+): (found/calcd)
1	**2a**, **2a′**	CH_3−	(3-NO_2)Ph−	0.72/1.00 (40:60): *S*/*R*	317.7/318.0
2	**2b**, **2b′**	$(CH_3)_2CH$−	(4-OMe)Ph−	1.42/1.58 (10:90): *R*/*S*	330.7/330.1
3	**2c**, **2c′**	CH_3−	(2,5-Me)Ph−	1.27/1.44 (62:38): *R*/*S*	300.7/301.1
4	**2d**	H−	furyl−	0.21(N/A): (N/A)	248.8/249.0
5	**2e**, **2e′**	$(CH_3)_2CHCH_2$−	Ph−	1.64/1.89 (06:94): *S*/*R*	314.9/315.4
6	**2f**, **2f′**	$PhCH_2$−	(3-F)Ph−	2.16/2.69 (18:82): n.d.[d]	366.7/367.1
7	**2g**, **2g′**	$PhCH_2$−	Ph−	2.03/2.43 (08:92): n.d.[d]	348.6/349.1

[a] Retention time in minutes. HPLC chromatograms were run with a gradient of 5−95% acetonitrile in water (0.05% TFA) over 6 min at 214 nm. [b] Configuration of the carbon *C-3 was determined by NOE experiments (see the Supporting Information). [c] Diastereoisomeric ratios were determined on the basis of the integration of the HPLC traces. [d] Not determined. In the case of phenylalanine derivatives, configurations could not be clearly assigned.

Table 2. Formation of Compounds 2 from Resin-Bound Primary Amines 1

entry	compds	R_1	R_2	mass (M + H^+) (found/calcd)	yield[a] (%)	purity[b] (%)
1	**2e**, **2e′**	$(CH_3)_2CHCH_2$−	Ph−	315.1/315.1	83	79
2	**2g**, **2g′**	$PhCH_2$−	Ph−	349.5/349.1	76	68
3	**2h**, **2h′**	$(CH_3)_2CH$−	Ph−	301.0/301.1	82	83
4	**2i**, **2i′**	CH_3−	Ph−	273.6/273.1	83	80
5	**2j**	H−	Ph−	259.7/259.1	98	75

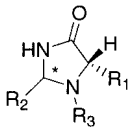
[a] Crude yields were calculated on the basis of the initial loading of the resin. [b] Purities were determined by integration of the HPLC traces at 214 nm. Chromatograms were run with a gradient of 5−95% acetonitrile in water (0.05% TFA) over 30 min at 214 nm.

in situ from ethyl bromodifluoroacetate, trimethylsilyl chloride, and zinc. Application of this reaction to additional substrates other than amino acids is in progress. These derivatives represent a useful tool for the generation of large libraries of fluorinated compounds including peptides, β-peptides, and peptidomimetics. Furthermore, the presence of the carboxylic acid group can be explored for a wide range of other possible transformations.

Experimental Section

General Methods. MBHA resin, 1% divinylbenzene, 100−200 mesh, 1.10 mequiv/g substitution, and *N*,*N*′-diisopropylcar-

Table 3. Formation of Imidazolidinones 11 from Resin-Bound Secondary Amines 9

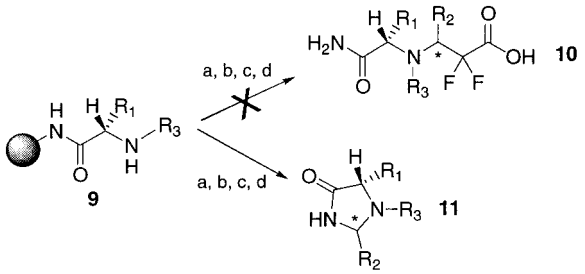


11

entry	compds	R_1	R_3	R_2	t_R[a] (**11x**/**11x′**) (ratio)[b]	mass ($M + H^+$): (found/calcd)	yield[c] (%)
1	**11a**	$-CH_2CH_2CH_2-$		Ph−	0.50 (N/A)	202.6/203.1	95
2	**11b**, **11b′**	$(CH_3)_2CHCH_2-$	CH_3	Ph−	1.94/2.10 (50:50)	232.6/233.1	90
3	**11c**, **11c′**	$PhCH_2-$	CH_3	Ph−	2.42/3.14 (50:50)	266.6/267.1	91

[a] Retention time in minutes. HPLC chromatograms were run with a gradient of 5−95% acetonitrile in water (0.05% TFA) over 6 min at 214 nm. [b] Diastereoisomeric ratios were determined on the basis of the integration of the HPLC traces. [c] Crude yields were calculated on the basis of the initial loading of the resin.

Scheme 4[a]



[a] Key: (a) BtH, R_2CHO, benzene, reflux; (b) Reformatsky reaction; (c) $LiOH·H_2O$; (d) HF/anisole.

bodiimide (DIC) were purchased from Chem Impex International (Wood Dale, IL). Boc-amino acids and *N*-hydroxybenzotriazole (HOBt) were purchased from Calbiochem-Novabiochem Corp. (San Diego, CA) and Bachem Bioscience Inc. (Philadelphia, PA). HF was purchased from Air Products (San Marcos, CA). Ethyl bromodifluoroacetate was purchased from Fluorochem USA (West Columbia, SC). All other reagents and anhydrous solvents were purchased from Aldrich Chemical Co. (Milwaukee, WI). The ^1H, ^{13}C, 2D-COSY, HMBC, HMQC, and NOE difference ^1D spectra were recorded in DMSO-d_6 solutions at 360 (^1H) and 90 (^{13}C) MHz, respectively. The ^{19}F spectra were recorded in DMSO-d_6 solutions at 470 MHz. The yields for purified products were determined on the basis of the theoretical loading of the polymeric support and starting from 100 mg of the resin.

Typical Procedure for the Synthesis of Compounds 2. *p*-Methylbenzhydrylamine (MBHA) resin (100 mg, 1.10 mequiv/g, 100−200 mesh) was sealed inside a polypropylene mesh packet. Polypropylene bottles were used for all the reactions. The resin was washed with dichloromethane (DCM) followed by neutralization with 5% diisopropylethylamine (DIEA) in DCM and washed with DCM.

(1) Coupling of an Amino Acid to the Resin. Boc-amino acid (0.66 mmol) was coupled to MBHA resin (0.11 mmol) using the classical coupling reagents DIC and HOBt (0.66 mmol) in anhydrous DMF (0.1 M) for 2 h at room temperature followed by washes with DMF (three times) and DCM (three times). The Boc group was deprotected using 55% TFA in DCM for 30 min followed by neutralization with 5% DIEA in DCM. Completeness of the coupling was verified by the ninhydrin test.

(2) Preparation of *N*-(α-Aminoalkyl)benzotriazoles 3. In a nitrogen-flushed reaction tube were mixed benzotriazole (2.62 g, 22 mmol), the resin-bound amino acids **1** (10 "tea bags", 1.1 mmol), and the aldehyde (22 mmol) in dry benzene (80 mL). The mixture was heated under reflux with a Dean−Stark trap during 3 h. Following washes with DCM, the resin was dried in vacuo and used in the Reformatsky coupling.

(3) Preparation of α,α-Difluoro-*β*-amino Esters 4. To a refluxing solution of chlorotrimethylsilane 1 M in THF (11 mL, 11 mmol) in dry THF (80 mL) was added Zn (720 mg, 11 mmol). The mixture was vigorously stirred, and ethyl bromodifluoroacetate (1.410 mL, 11 mmol) was added dropwise, followed by the resin-bound benzotriazolyl derivatives **3** (10 "tea bags", 1.1 mmol). After 2 h of refluxing, the resin was washed with DCM (three times), DMF (three times), and DCM (three times).

(4) Preparation of α,α-Difluoro-*β*-amino Acids 2. The resin-bound ethyl esters **4** (10 "tea bags", 1.1 mmol) were treated overnight with $LiOH·H_2O$ (460 mg, 11 mmol) in solution in H_2O/THF 1:5 (0.1 M). The resin was washed with DCM (three times), DMF (three times), and DCM (three times). The resin was cleaved by anhydrous HF in the presence of anisole at 0 °C for 1.5 h, and the cleaved product was extracted with 95% acetic acid in H_2O and lyophilized.

3-(1′(*S*)-Carbamoyl-3′-methylbutylamino)-2,2-difluoro-3(*S*)-phenylpropionic Acid (2e). A total of 3 mg was obtained following purification with preparative HPLC (yield: 9%). ^1H NMR (DMSO-d_6, δ ppm, 360 MHz): 0.83 (d, 3H, J = 6.6 Hz), 0.86 (d, 3H, J = 6.6 Hz), 1.30 (ddd, 1H, J = 13.4, 5.6, 7.9 Hz), 1.42 (ddd, 1H, J = 13.4, 8.1, 5.7 Hz), 1.73 (m, 1H, J = 6.6, 5.7, 7.9 Hz), 3.17 (dd, 1H, J = 8.1, 5.6 Hz), 4.34 (dd, 1H, J = 19.7, 7.4 Hz), 6.91 (s, 1H), 7.32 (s, 2H), 7.39−7.30 (m, 5H). ^{13}C NMR (DMSO-d_6, δ ppm, 90 MHz): 22.8, 22.1, 23.7, 42.1, 58.6, 62.8 (dd, J = 23.7, 25.6 Hz), 114.4 (t, J = 264.1 Hz), 128.0, 128.4, 128.9, 133.8, 166.2 (t, J = 27.3 Hz), 174.4. NOE diff ^1D: irradiation at 7.35 ppm (H-2″, H-3″, H-4″, H-5″, H-6″), no NO effect for H-5′ and H-4′. ^{19}F NMR (DMSO-d_6, δ ppm, 470 MHz): −105.18 (d, J = −319.8 Hz), −116.44 (d, J = −319.8 Hz). MS (ESI): calcd [MH^+] 315.1, found 314.9.

3-(1′(*S*)-Carbamoyl-3′-methylbutylamino)-2,2-difluoro-3(*R*)-phenylpropionic Acid (2e′). A total of 15 mg was obtained following purification with preparative HPLC (yield: 43%). ^1H NMR (DMSO-d_6, δ ppm, 360 MHz): 0.61 (d, 3H, J = 6.6 Hz), 0.76 (d, 3H, J = 6.6 Hz), 1.30 (AB, 1H, J = 5.8, 8.0, 13.6 Hz), 1.39 (AB, 1H, J = 7.9, 5.8, 13.6 Hz), 1.66 (m, 1H, J = 6.6, 5.8, 8.0 Hz), 2.81 (dd, 1H, J = 7.9, 5.8 Hz), 4.22 (dd, 1H, J = 14.6, 12.2 Hz), 7.18 (s, 1H), 7.21 (s, 1H), 7.36−7.45 (m, 5H). ^{13}C NMR (DMSO-d_6, δ ppm, 90 MHz): 21.8, 22.8, 23.9, 42.1, 57.4, 62.5 (t, J = 23.7 Hz), 114.3 (t, J = 255.1 Hz), 128.3, 128.8, 129.3, 133.2, 164.2 (t, J = 30.3 Hz), 174.4; NOE diff ^1D: irradiation at 7.35 ppm (H-2″, H-3″, H-4″, H-5″, H-6″), NO effect for H-5′ and H-4′. ^{19}F NMR (DMSO-d_6, δ ppm, 470 MHz): −110.00 (d, J = −315.1 Hz), −113.15 (d, J = −315.1 Hz). MS (ESI): calcd [MH^+] 315.1, found 314.7.

3-Phenyl-2-(2′,2′,4′,4′-tetrafluoro-3′,3′-dihydroxy-1′-phenylbutylamino)propionamide (8g). A total of 17 mg was obtained following purification with preparative HPLC (yield: 38%). ^1H NMR (DMSO-d_6, 360 MHz): δ 2.70 (dd,1H, J = 13.6, 8.1 Hz), 2.91 (dd, 1H, J = 13.6, 5.1 Hz), 3.11 (dd, 1H, J = 5.1, 8.1 Hz), 4.25 (dd, 1H, J = 24.8, 2.6 Hz), 6.02 (t, 1H, J = 54.2 Hz), 7.08 (d, 2H, J = 7.1 Hz), 7.11 (dd, 2H, J = 7.8, 2.1 Hz), 7.19−7.27 (m, 2H), 7.22−7.30 (m, 2H), 7.19−7.33 (m, 1H), 7.22−7.33 (m, 1H), 7.38 (s, 1H), 7.63 (s, 1H). ^{13}C NMR (DMSO-d_6, 90 MHz): δ 39.0, 59.6, 60.5 (dd, J = 20.5, 28.6 Hz), 92.3 (t, J = 27.3 Hz), 112.6 (t, J = 248.8 Hz), 115.5 (t, J = 253.4 Hz), 126.4, 127.9, 128.1, 128.2, 129.0, 129.1, 134.1, 137.2, 174.5. ^{19}F NMR (DMSO-d_6, 470 MHz): δ −110.3 (dt, J = −315.1, −9.4 Hz); −112.97 (ddd, J = −315.1, −18.8, −9.4 Hz); −122.86 (dm, J = −324.5 Hz); −124.42 (dm, J = −329.2 Hz); −128.48 (ddd, J = −357.5, −14.1 Hz, −9.4 Hz); −132.43 (dt, J = −357.5, −11.7 Hz); −134.14 (dt, J = −357.5, -11.7 Hz); −134.57 (ddd, J = −357.5, −18.8, −9.4 Hz). MS (ESI): calcd [MH^+] 401.1, found 400.7.

3-Phenylhexahydropyrrolo[1,2-*c*]imidazol-1-one (11a). A total of 16 mg was obtained following purification with preparative HPLC (yield: 72%). ^{1}H NMR (DMSO-d_6, 360 MHz): δ 2.0−1.84 (m, 2H), 2.18−2.03 (m, 2H), 3.56−3.38 (m, 2H), 4.53−4.33 (m, 1H), 5.76 (s, 1H), 7.51−7.47 (m, 2H), 7.57−7.51 (m, 3H), 9.34 (s, 1H). ^{13}C NMR (DMSO-d_6, 90 MHz): δ 24.0, 26.9, 55.9, 62.8, 77.0, 127.1, 128.8, 129.9, 136.8, 171.5. MS (ESI): calcd [MH^+] 203.1, found 202.6.

Acknowledgment. This work was supported by National Cancer Institute Grant No. CA 78040 (R.A.H.). We thank Dr. Jean-Pierre Vidal for helpful discussion.

Supporting Information Available: Copies of ^{1}H, ^{13}C, COSY, HMBC, HMQC, NOE difference ^{1}D and ^{19}F NMR spectra of **2e**, **2e′**, **8g**, and **11a**. Description of the other compounds: **2a**, **2a′**, **2b**, **2b′**, **2c**, **2c′**, **2d**, **2f**, **2f′**, **2g**, **2g′**, **11b**, **11b′**, and **11c**, **11c′**. This material is available free of charge via the Internet at http://pubs.acs.org.

JO010872Z