Synthetic Communications

ISSN: 0039-7911 (Print) 1532-2432 (Online) Journal homepage: http://www.tandfonline.com/loi/lsyc20

Efficient Synthesis of O-Succinimidyl-(tert-Butoxycarbonylamino)methyl Carbamates Derived from α-Amino Acids Accelerated by Ultrasound: Application to the Synthesis of Ureidodipeptides

Vommina V. Sureshbabu , Naremaddepalli S. Sudarshan & Kantharaju

To cite this article: Vommina V. Sureshbabu , Naremaddepalli S. Sudarshan & Kantharaju (2008) Efficient Synthesis of O-Succinimidyl-(tert-Butoxycarbonylamino)methyl Carbamates Derived from α-Amino Acids Accelerated by Ultrasound: Application to the Synthesis of Ureidodipeptides, Synthetic Communications, 38:13, 2168-2184, DOI: 10.1080/00397910802026386

To link to this article: http://dx.doi.org/10.1080/00397910802026386

Published online: 18 Aug 2008.

Synthetic Communications®, 38: 2168–2184, 2008

ISSN: 0039-7911 print/1532-2432 online
DOI: 10.1080/00397910802026386

Taylor & Francis
Taylor & Francis Group

Efficient Synthesis of *O*-Succinimidyl-(*tert*-Butoxycarbonylamino)methyl Carbamates Derived from α-Amino Acids Accelerated by Ultrasound: Application to the Synthesis of Ureidodipeptides

Vommina V. Sureshbabu, Naremaddepalli S. Sudarshan, and Kantharaju
Department of Studies in Chemistry, Central College Campus, Bangalore University, Bangalore, India

Abstract: The synthesis of *O*-succinimidyl-(*tert*-butoxycarbonylamino)methyl carbamates employing isocyanates made through the Curtius rearrangement of Boc-amino acid azides in the presence of *N*-hydroxysuccinimide under the influence of ultrasound is described.

Keywords: Boc-amino acid azide; Boc-amino acid isocyanate; *N*-hydroxysuccinimide; Ultrasonic irradiation

INTRODUCTION

Ureidopeptides are an important class of peptidomimetics by virtue of their applications as drugs with more elevated biological properties than the parent peptides[1,2] as well as structural motifs for conformational analysis.[3,4] Therefore, development of new methods to synthesize these compounds has gained attention. Recently, Fishcer et al. have reported the synthesis of six *O*-succinimidyl-(*tert*-butoxycarbonylamino)methyl carbamates[5] through the Curtius rearrangement of corresponding acid azides under high-temperature reflux. In this case, the product yields have

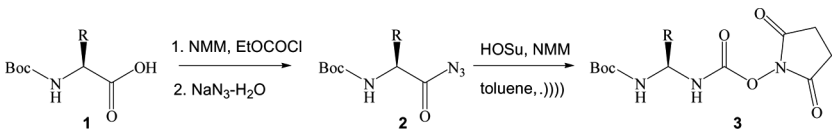
Received August 20, 2007
Address correspondence to Vommina V. Sureshbabu, Department of Studies in Chemistry, Central College Campus, Dr B. R. Ambedkar Veedhi, Bangalore University, Bangalore 560 001, India. E-mail: hariccb@rediffmail.com

shown to be less then satisfactory, presumably because of the decomposition of Boc-protected derivatives at the elevated temperature used for the reaction. Consequently, the preparation of isocyanates derived from Boc amino acids requires an alternative strategy. We herein demonstrate the Curtius rearrangement of Boc amino acid azides under ultrasonication at ambient temperature.

RESULTS AND DISCUSSION

Boc-/Z-amino acid azides are known for their instability even at room temperature.[6] Their utility as peptide coupling agents at ambient temperature was accompanied by the formation of the reactive isocyanates leading to several side products. Recently, Guichard et al. reported an efficient stepwise synthesis of *N,N′*-linked oligoureas [HN-CHR-CH_2-NH-CO]$_n$ in solution and on solid support utilizing *O*-succinimidyl carbamates derived from *N*-Boc-/Fmoc-β^3-amino acids as well as Boc-/Z-/Fmoc-α -amino acids and dipeptides as activated monomers.[7,8] Further, they have explained the short-range interaction of the *cis–trans* urea motif in urea peptides Boc-*L*-*g*Ser(Bzl)-CONHMe/NMe_2, Piv-*L*-Pro-*L*-gLeu-CONHMe/NMe_2, Boc-L-*g*Ser(Bzl)-CO-*L*-Leu-NHMe, and Boc-L-*g*Ser(Bzl)-CO-*D*-Leu-NMe_2.[9] Subsequently, they have also described solution-phase synthesis of hexahydro-1,3,5-triazepine-2,6-diones, novel rigid, highly substituted, seven-membered ring urea-based scaffolds.[10]

In the present study, the Boc-α -amino acid **1** (1 equiv) was dissolved in THF (5 mL) and cooled to -15°C. After the addition of ethyl chloroformate (EtOCOCl, 1.1 equiv) and *N*-methylmorpholine (NMM, 1.1 equiv), the mixture was stirred at -15°C for 20 min. The resulting white suspension was allowed to warm to -5°C, treated with an aqueous solution (1 mL) of NaN_3 (2.5 equiv), and stirred for 10 min. After usual aqueous workup, the resulting Boc-α -amino acid azide in toluene was exposed to ultrasound at ambient temperature in the presence of an equimolar quantity of *N*-hydroxysuccinimide and NMM. Initially they rearranged to the corresponding isocyanates, which are trapped by

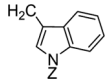
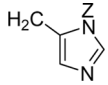


Scheme 1. Synthesis of *O*-Succinimidyl(Boc)methylcarbamates.

N-hydroxysuccinimide, giving the desired *O*-succinimidyl-(*tert*-butoxycarbonylamino) methyl carbamate **3a–t** (Scheme 1) in high yields.

The entire course of the reaction was complete in about 15–20 min. The methyl carbamates **3** precipitated directly from the toluene solution were simply collected by filtration and washed with toluene. All the methyl carbamates **3** were characterized by IR, ^{1}H, ^{13}C NMR, and mass spectrometry. Their IR spectrum contains a characteristic sharp peak of methyl carbamate urethane stretching vibrational frequency at around

Table 1. Physical constants of O-succinimdyl-(tert-butoxycarbonylamino)methyl carbamates

Compound **3**	R	R^1	Yield (%)	Mp (°C)	MALDI mass: Calculated $[M]^+$	MALDI mass: Found $[M + Na]^+$
a	H	CH_3	87	152	301.3	324.2
b	H	H	82	166	287.3	310.2
c	H	$CH(CH_3)_2$	85	107	329.4	352.3
d	H	$CH_2CH(CH_3)_2$	84	125	343.4	366.3
e	H	$CH(CH_3)C_2H_5$	82	129	343.4	366.2
f	H	$CH_2C_6H_5$	86	143	377.4	400.3
g	H	C_6H_5	87	126	363.4	386.3
h*	H	C_6H_5	85	122	363.4	386.3
i	H	CH_2OBzl	83	141	407.4	430.2
j	H	$CH_2COOBzl$	84	122	435.4	458.5
k	H	$(CH_2)_2COOBzl$	80	140	449.5	472.2
l	$R = R^1$	$-(CH_2)_3-$	82	127	327.3	350.3
m	H	$(CH_2)_2SCH_3$	80	125	347.4	370.3
n	H	$(CH_2)_4NHZ$	75	149	492.5	515.2
o	H	$CH_2C_6H_5OH$	72	151	393.4	416.3
p	H	$CH(CH_3)OH$	76	142	331.3	354.3
q	H		70	168	550.5	573.2
r	H	$(CH_2)_2CONH_2$	72	136	358.3	381.3
s	H	CH_2CONH_2	75	141	344.3	367.1
t	H		70	149	501.4	524.3

Notes: Phg = phenylglycine; *D-configuration.

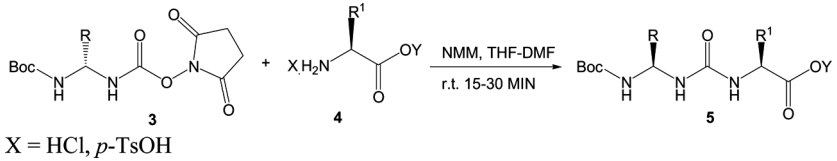
Table 2. Comparison of Boc-gPhe-COOSu (3f) formation under different methods

Methods	Time (min)	Yield (%)
Thermal	45	43
Microwave	2	49
Ultrasound	25	86

1735–1745 cm^{-1}. The physical constants as well as yields 70–85% of methyl carbamates are given in the Table 1. Almost all the proteniogenic amino acids including Ser, Thr, Tyr, Asp, Glu, Asn, Gln, His, Trp, and Cys have been prepared. The methyl carbamates **3** were stable white crystalline solids. When stored at rt for long periods, neither noticeable degradation nor any change in their spectral behavior was observed. A comparative study for the synthesis of *O*-succinimidyl-{1-(*tert*-butoxycarbonylamino)-2-phenyl}methylcarbamate using Boc-Phe-N_3 and N-hydroxysuccinimide was carried out under different conditions. It was found that the formation of **3** through Curtius rearrangement assisted by ultrasound under ambient temperature results in better yields then the classical thermal method or microwave irradiation, and results are furnished in Table 2.

To demonstrate the use of the activated carbamates, the synthesis of Boc-dipeptidyl urea esters has been carried out. The reaction of methylcarbamate **3** with amino acid esters **4** in THF–DMF (2:1) in the presence of NMM at rt gave the corresponding ureas **5** in good yields (Scheme 2). The reaction proceeds rapidly and was found to be completed within 30 min at rt. A routine workup and recrystallization with DMSO–water resulted in analytically pure dipeptidyl urea esters **5a–j**. The only by-product formed during the reaction was *N*-hydroxysuccinimide, which was easily removed during aqueous wash. All the dipeptidyl ureas prepared are characterized by ^1H and ^{13}C NMR and mass spectral methods.

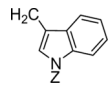
The carbamates were further reacted with the in situ generated *N, O*-bis(trimethylsilyl)amino acids **7** to synthesize Boc-dipeptidyl urea acids



X = HCl, *p*-TsOH

Scheme 2. Synthesis of ureidopeptide esters from **3**.

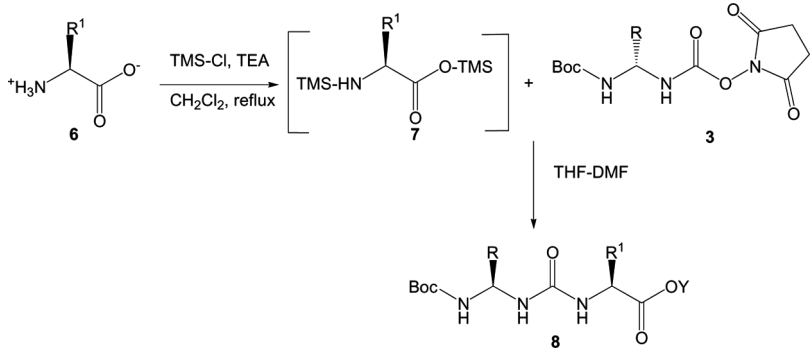
Table 3. List of ureidopeptide esters prepared through scheme 2

Urea ester 5	R	R^1	Y
a	CH(CH3)2	CH3	CH3
b	CH2C6H5	CH(CH3)2	CH3
c	H	CH2C6H5	CH3
d	CH2C6H5	CH3	CH3
e	CH2OBzl	H	CH3
f	CH2COOBzl	H	CH3
g	(CH2)2COOBzl	CH2CH(CH3)2	CH3
h	CH(CH3)C2H5	CH2C6H5	CH3
i	CH2C6H4OH	CH3	CH3
j		CH(CH3)2	CH3

(Scheme 3). A simple workup of the reaction mixture followed by their recrystallization resulted in pure dipeptidyl urea acids **8a–d**, which were fully characterized.

Racemization Study for Methylcarbamates 3

To demonstrate the stereospecificity of both the carbamates **3** prepared and their use as urea coupling agents, the carbamate **3f** derived from Boc-Phe was coupled with R-(+), S-(−), and R,S-(±)-1-phenylethylamines **9a–c** to obtain the white solid ureas **10a**, **10b**, and **10c** respectively (Scheme 4, Figure 1), and their recrystallization resulted in analytically



Scheme 3. Synthesis of ureidopeptide acids **8** (Y-H) from **3**.

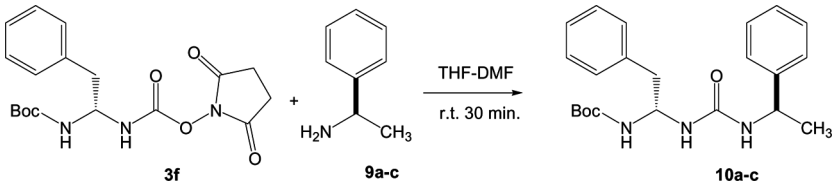
Table 4. List of ureidopeptide acids obtained via scheme 3

Urea acid 8	R	R^1
a	$CH_2CH(CH_3)_2$	CH_3
b	$CH_2C_6H_5$	CH_3
c	$CH_2CH(CH_3)_2$	H
d	$CH(CH_3)_2$	$CH_2C_6H_5$

pure compounds. Their ^{1}H NMR analysis revealed that the methyl group resonances of **10a** (δ, 1.28 and 1.30) and **10b** (δ, 1.26 and 1.28) are clearly separated by 0.02 ppm in DMSO-d_6 solution. In addition, the ^{1}H NMR spectrum of the racemic urea **10c** was observed as a doublet of doublet for the methyl group resonances at (δ, 1.25, 1.27, 1.28, and 1.30). This clearly confirms that both the carbamates' preparation as well as the coupling reaction to obtain the peptidyl ureas are completely free from racemization.

EXPERIMENTAL

The melting points were recorded in open capillary tubes and are uncorrected. The reactions were carried out using a sonic bath (35 kHz, Elma, T 310/H German make) at ambient temperature. Infrared spectra were recorded on a Nicolet Impact 400D FT-IR spectrometer (KBr pellets, 3 cm^{-1} resolution). Elementalanalyses were carried out using a Perkin-Elmer analyzer, and the samples were dried for 24 h under vacuum before analysis. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AMX 400-MHz spectrometer. Mass spectra were recorded on Maldi, PE-Sciex 150 EX LC-MS, and Jeol-JMS-600H spectrometers in FAB^+ mode using 3-nitrobenzyl alcohol as a liquid matrix. All solvents were freshly distilled prior to use. Amino acid methyl ester hydrochlorides were prepared using methanol and thionyl chloride.



Scheme 4. Preparation of urea adducts for racemization studies.

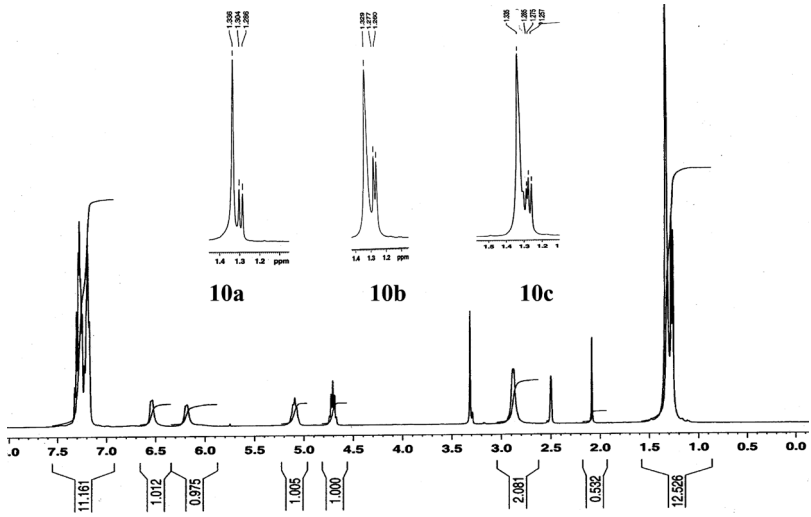


Figure 1. ^{1}H NMR spectra of Boc-Phe-ψ(NH-CO-NH)-R-(+)-1-phenylethylamine: Inset figure shown splitting pattern of CH3 group of 1-phenylethylamine in urea adducts R-(+)-1-phenylethylamine (**10a**), S-(−)-1-phenylethylamine (**10b**), and R,S-(±)-1-phenylethylamine (**10c**).

General Procedure for the Synthesis of *O*-Succinimidyl-(*tert*-butoxycarbonylamino)methylcarbamates 3a–t

Boc-α-amino acid **1** (1 mmol) was dissolved in THF (5 mL) and cooled to –20 °C. After addition of ethylchloroformate (1.1 mmol) and *N*-methylmorpholine (1.1 mmol), the mixture was stirred at –20 °C for 15 min and allowed to warm to − 5°C. It was treated with an aqueous solution of NaN_3 (2.5 mmol in 2 mL of water) for 5 min. After concentration of the solvent under reduced pressure, the resulting residue was diluted with CH_2Cl_2 (15 mL); washed with 5% citric acid solution (3 × 5 mL), 5% sodium bicarbonate solution (3 × 5 mL), and brine; dried over anhydrous Na_2SO_4; and concentrated under reduced pressure to give the acyl azide **2**, which was used without further purification. The acyl azide was taken in toluene (5 mL), and *N*-hydroxy succinimide (1 mmol) and NMM (1.1 mmol) were successively added. The mixture was sonicated in an ultrasound bath for about 15–25 min at ambient temperature. After the completion of the reaction, carbamates crystallized from the toluene solution were collected by filtration. All the carbamates **3** have been fully characterized. The physical and spectral data are furnished in Table 1.

Data

Boc-gAla-COOSu **(3a)**

White solid,IR: 1715, 1745, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.16 (3H, s), 1.36 (9H, s), 2.8 (4H, s), 3.8 (1H, m), 5.2 (1H, m), 8.5 (1H, d); ^{13}C NMR (DMSO): δ 18.3, 28.6, 59.6, 70.1, 72.2, 78.4, 151.2, 154.7, 170.8. Anal. calcd. for $C_{12}H_{19}N_3O_6$: C, 47.84; H, 6.36; N, 13.95. Found: C, 47.12; H, 6.62; N, 13.24%.

Boc-gGly-COOSu **(3b)**

White solid,IR: 1710, 1750, 1775 cm^{-1}; ^1H NMR (DMSO): δ 1.36 (9H, s), 2.5 (2H, m), 2.8 (4H, s), 5.2 (1H, br), 8.2 (1H, br); ^{13}C NMR (DMSO): δ 27.8, 40.4, 69.6, 72.0, 79.1, 151.2, 155.7, 170.5. Anal. calcd. for $C_{11}H_{17}N_3O_6$: C, 45.99; H, 5.96; N, 14.63. Found: C, 45.12; H, 5.62; N, 14.50%.

Boc-gVal-COOSu **(3c)**

White solid,IR: 1705, 1745, 1770 cm^{-1}; ^1H NMR (DMSO): δ 0.93 (6H, t), 1.35 (9H, s), 1.9 (1H, m), 2.7 (4H, s), 3.9 (1H, m), 5.3 (1H, br), 8.4 (1H, d); ^{13}C NMR (DMSO): δ 18.5, 19.7, 28.4, 29.6, 58.6, 69.7, 72.1, 79.0, 150.7, 155.2, 170.8. Anal. Calcd. for $C_{14}H_{23}N_3O_6$: C, 51.06; H, 7.04; N, 12.76. Found: C, 51.12; H, 7.62; N, 12.24%.

Boc-gLeu-COOSu **(3d)**

White solid,IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 0.93 (6H, d), 1.25–1.35 (11H, m), 1.62 (1H, m), 2.7 (4H, s), 3.7 (1H, m), 5.2 (1H, br), 8.5 (1H, br); ^{13}C NMR (DMSO): δ 22.1, 23.4, 24.8, 41.0, 51.6, 70.1, 72.2, 78.7, 151.2, 154.7, 171.0. Anal. calcd. for $C_{15}H_{25}N_3O_6$: C, 52.47; H, 7.34; N, 12.24. Found: C, 52.12; H, 7.62; N, 12.50%.

Boc-gIle-COOSu **(3e)**

White solid,IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 0.91 (6H, m), 1.12 (1H, m), 1.3 (9H, s), 1.52 (2H, m), 3.7 (1H, m), 4.94 (1H, m), 5.2 (1H, br); ^{13}C NMR (DMSO): δ 11.5, 15.7, 25.2, 26.2, 28.6, 34.4, 56.6, 78.2, 151.2, 153.7, 170.5. Anal. calcd. for $C_{15}H_{25}N_3O_6$: C, 52.47; H, 7.34; N, 12.24. Found: C, 53.12; H, 7.0; N, 12.10%.

Boc-gPhe-COOSu **(3f)**

White solid, IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.36 (9H, m), 2.7 (4H, s), 2.8 (2H, d), 3.9 (1H, m), 5.0 (1H, d), 5.4 (1H, m), 7.25–7.3 (5H, m); ^{13}C NMR (DMSO): δ 26.1, 28.9, 62.2, 79.1, 126.9, 127.6, 128.9, 129.0, 129.1, 130.2, 137.9, 151.4, 155.2, 171.6. Anal. calcd. for $C_{18}H_{23}N_3O_6$: C, 57.29; H, 6.14; N, 11.13. Found: C, 57.12; H, 6.20; N, 11.24%.

Boc-gPhg-COOSu **(3g)**

White solid, IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.3 (9H, s), 2.68 (4H, s), 3.9 (1H, m), 4.9 (1H, d), 5.2 (1H, br), 7.25–7.3 (5H, m); ^{13}C NMR (DMSO): δ 26.6, 28.6, 57.8, 78.2, 125.8, 127.1, 127.9, 129.7, 151.2, 154.7, 171.5. Anal. calcd. for $C_{17}H_{21}N_3O_6$: C, 56.19; H, 5.83; N, 11.56. Found: C, 56.12; H, 5.62; N, 11.24%.

Boc-g(D)Phg-COOSu **(3h*)**

White solid, IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.28 (9H, m), 2.7 (4H, s), 3.88 (1H, m), 4.8 (1H, m), 5.1 (1H, br), 7.25–7.3 (5H, m); ^{13}C NMR (DMSO): δ 26.7, 28.8, 58.6, 79.0, 125.6, 127.1, 127.9, 129.7, 151.8, 155.7, 170.9. Anal. calcd. for $C_{17}H_{21}N_3O_6$: C, 56.19; H, 5.83; N, 11.56. Found: C, 56.10; H, 5.66; N, 11.20%.

Boc-gSer(Bzl)-COOSu **(3i)**

White solid, IR: 1715, 1755, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.3 (9H, s), 3.6 (2H, d), 3.8 (1H, m), 4.8 (2H, s), 5.1 (1H, d), 5.3 (1H, br), 7.25–7.30 (5H, m); ^{13}C NMR (DMSO): δ 25.6, 27.8, 51.4, 62.6, 78.2, 124.5, 125.1, 127.1, 127.9, 151.2, 153.9, 171.5. Anal. calcd. for $C_{19}H_{25}N_3O_7$: C, 56.01; H, 6.18; N, 10.31. Found: C, 56.10; H, 6.62; N, 10.70%.

Boc-gAsp(Bzl)-COOSu **(3j)**

White solid, IR: 1715, 1755, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.3 (9H, s), 3.6 (2H, d), 3.8 (1H, m), 4.0 (2H, s), 4.9 (1H, d), 5.1 (1H, m), 7.25–7.35 (5H, m); ^{13}C NMR (DMSO): δ 26.0, 28.6, 37.2, 48.4, 57.6, 78.6, 125.1, 127.1, 127.9, 129.7, 151.2, 153.7, 154.8, 170.5. Anal. calcd. for $C_{20}H_{25}N_3O_8$: C, 59.19; H, 4.59; N, 5.11. Found: C, 59.12; H, 4.62; N, 5.24%.

Boc-gGlu(Bzl)-COOSu **(3k)**

White solid, IR: 1715, 1755, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.32 (9H, s), 3.6–3.8 (5H, m), 4.9 (2H, s), 5.1 (1H, d), 5.3 (1H, m), 7.25–7.35 (5H, m); ^{13}C NMR (DMSO): δ 25.8, 28.2, 35.6, 37.5, 48.8, 56.9, 78.6, 125.1, 127.1, 127.9, 129.7, 151.2, 153.7, 154.8, 171.5. Anal. calcd. for $C_{21}H_{27}N_3O_8$: C, 56.12; H, 6.06; N, 9.35. Found: C, 56.00; H, 6.62; N, 9.24%.

Boc-gPro-COOSu **(3l)**

White solid, IR: 1710, 1745, 1780 cm^{-1}; ^1H NMR (DMSO): δ 1.2–2.2 (13H, m), 2.7 (4H, s), 3.4–3.7 (3H, m), 4.8 (1H, d), 5.1 (1H, m); ^{13}C NMR (DMSO): δ 24.2, 25.8, 28.2, 28.4, 60.6, 78.1, 150.8, 154.7, 170.4. Anal. calcd. for $C_{14}H_{21}N_3O_6$: C, 51.37; H, 6.47; N, 12.84. Found: C, 51.12; H, 6.62; N, 12.24%.

Boc-gMet-COOSu **(3m)**

White solid, IR: 1710, 1750, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.31–2.25 (11H, m), 2.7 (4H, s), 3.3–3.5 (4H, m), 4.9 (1H, d), 5.15 (1H, m); ^{13}C NMR (DMSO): δ 26.1, 27.9, 28.5, 48.5, 49.1, 59.8, 73.9, 151.8, 154.7, 171.4. Anal. calcd. for $C_{13}H_{21}N_3O_6S$: C, 44.95; H, 6.09; N, 12.09. Found: C, 45.12; H, 6.02; N, 12.06%.

Boc-gLys(Z)-COOSu **(3n)**

White solid, IR: 1715, 1750, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.37 (9H, s), 1.6 (6H, m), 3.0 (6H, m), 4.8 (2H, s), 5.0 (1H, s), 5.4 (1H, m), 5.8 (1H, m), 7.25–7.30 (5H, m); ^{13}C NMR (DMSO): δ 18.9, 24.1, 25.3, 27.8, 32.1, 47.4, 59.7, 69.0, 79.4, 127.7, 128.4, 137.4, 152.4, 156.6, 158.3, 172.9. Anal. calcd. for $C_{23}H_{32}N_4O_8$: C, 56.09; H, 6.55; N, 11.38; O, 25.99. Found: C, 56.01; H, 6.52; N, 11.30; O, 25.91%.

Boc-gTyr-COOSu **(3o)**

White solid, IR: 1715, 1745, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.37 (9H, s), 3.0 (4H, m), 7.1 (4H, m), 5.8 (1H, m); ^{13}C NMR (DMSO): δ 26.1, 28.1, 40.1, 63.9, 81.1, 116.0, 129.3, 129.4, 152.3, 154.8, 158.2, 172.2. Anal. calcd. for $C_{18}H_{23}N_3O_7$: C, 54.96; H, 5.89; N, 10.68; O, 28.47. Found: C, 54.90; H, 5.81; N, 10.70; O, 28.51%.

Boc-gThr-COOSu **(3p)**

White solid, IR: 1710, 1745, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.2 (3H, s), 1.38 (9H, s), 3.0 (4H, m), 3.7 (1H, m), 5.2 (1H, m), 5.8 (1H, m); ^{13}C NMR (DMSO): δ 17.9, 26.1, 28.2, 68.6, 70.8, 81.1, 153.1, 158.2, 172.8. Anal. calcd. for $C_{13}H_{21}N_3O_7$: C, 47.13; H, 6.39; N, 12.68; O, 33.80. Found: C, 47.08; H, 6.42; N, 12.70; O, 33.73%.

Boc-gTrp(Z)-COOSu **(3q)**

White solid, IR: 1719, 1745, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.37 (9H, s), 3.0 (4H, m), 3.4 (2H, m), 5.6 (m, H), 7.0–7.9 (10H, m); ^{13}C NMR (DMSO): δ 25.5, 28.1, 33.5, 63.4, 64.6, 81.0, 108.9, 119.2, 122.2, 122.5, 124.8, 127.6, 128.5, 127.9, 137.7, 146.8, 148.1, 153.8, 159.3, 172.2. Anal. calcd. for $C_{28}H_{30}N_4O_8$: C, 61.08; H, 5.49; N, 10.18; O, 23.25. Found: C, 61.01; H, 5.50; N, 10.10; O, 23.33%.

Boc-gGln-COOSu **(3r)**

White solid, IR: 1710, 1750, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.36 (9H, s), 2.1 (2H, m), 2.4 (2H, t), 3.1 (4H, m), 5.0 (2H, s), 5.8 (m, 1H); ^{13}C NMR (DMSO): δ 16.1, 25.5, 28.1, 32.6, 62.4, 81.0, 152.4, 158.6, 172.2, 172.4. Anal. calcd. for $C_{14}H_{22}N_4O_7$: C, 46.92; H, 6.19; N, 15.63; O, 31.25. Found: C, 46.88; H, 6.23; N, 15.67; O, 31.27%.

Boc-gAsn-COOSu **(3s)**

White solid, IR: 1715, 1750, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.36 (9H, s), 3.0 (6H, m), 4.8 (1H, s), 5.0 (2H, s), 5.7 (m, 1H); ^{13}C NMR (DMSO): δ 25.4, 28.0, 42.2, 62.0, 81.0, 149.9, 155.9, 172.3, 174.4. Anal. calcd. for $C_{13}H_{20}N_4O_7$: C, 45.35; H, 5.85; N, 16.27; O, 32.53. Found: C, 45.30; H, 5.90; N, 16.30; O, 32.56%.

Boc-gHis(Z)-COOSu **(3t)**

White solid, IR: 1710, 1745, 1785 cm^{-1}; ^1H NMR (DMSO): δ 1.37 (9H, s), 3.1 (6H, m), 5.2 (2H, s), 7.0 (6H, m), 7.6 (m, 1H); ^{13}C NMR (DMSO): δ 25.4, 28.1, 31.4, 63.5, 64.6, 81.0, 107.8, 122.2, 127.88, 127.95, 128.4, 128.9, 138.1, 146.1, 152.0, 153.8, 160.1, 172.3. Anal. calcd. for $C_{23}H_{27}N_5O_8$: C, 55.09; H, 5.43; N, 13.97; O, 25.52. Found: C, 55.02; H, 5.50; N, 13.90; O, 25.55%.

General Procedure for the Synthesis of Boc-Dipeptidyl Urea Esters (5a–j)

To a stirred solution of amino acid methyl ester hydrochloride salt **4** (1.3 mmol) in THF–DMF (2:1, 5 mL), NMM (1.3 mmol) and *O*-succinimidyl methylcarbamate **3** (1 mmol) were successively added and stirred at rt until the completion of the reaction. After the completion of the reaction, the solvent was evaporated under reduced pressure, and the residue was triturated with water, filtered, and finally recrystallized with DMSO–water to afford the pure urea esters **5a–j** as crystalline off-white solids.

Data

Boc-Val-ψ(NH-CO-NH)-Ala-OMe **(5a)**

Yield: 77%; mp 170°C; ^{1}H NMR (DMSO): δ 0.93 (6H, t), 1.15 (3H, d), 1.35 (9H, s), 1.85 (1H, m), 3.6 (3H, s), 3.8–3.95 (2H, m), 5.15 (1H, m), 6.4 (1H, d), 6.5 (1H, d); ^{13}C NMR (DMSO): δ 18.5, 18.8, 28.6, 32.9, 48.3, 52.0, 60.6, 63.0, 78.0, 155.0, 156.6, 174.5; ESI MS: m/z 319.2 $[M+H]^+$. Anal. calcd. for $C_{14}H_{27}N_3O_5$: C, 52.98; H, 8.57; N, 13.24. Found: C, 53.20; H, 8.52; N, 13.38%.

Boc-Phe-ψ(NH-CO-NH)-Val-OMe **(5b)**

Yield: 72%; mp 140°C; ^{1}H NMR (DMSO): δ 0.93 (6H, t), 1.3 (9H, s), 1.8 (1H, m), 2.85 (2H, d), 3.6 (3H, s), 3.8–3.95 (2H, m), 5.2 (1H, m), 6.35 (1H, br), 6.45 (1H, d), 7.15–7.3 (5H, m); ^{13}C NMR (DMSO): δ 18.7, 19.5, 28.3, 29.5, 37.1, 54.7, 57.5, 63.1, 78.2, 126.9, 127.6, 129.1, 137.9, 155.3, 156.9, 174.5; ESI MS: *m/z* 406.7 $[M+H]^+$. Anal. calcd. for $C_{21}H_{31}N_3O_5$: C, 62.20; H, 7.71; N, 10.36. Found: C, 61.90; H, 7.52; N, 10.38%.

Boc-Gly-ψ(NH-CO-NH)-Phe-OMe **(5c)**

Yield: 74%; mp 120–22°C; ^{1}H NMR (DMSO): δ 1.35 (9H, s), 2.85 (2H, d), 3.6 (3H, s), 3.85 (3H, m), 5.2 (1H, m), 6.3 (1H, br), 6.5 (1H, d), 7.2–7.4 (5H, m); ^{13}C NMR (DMSO): δ 28.7, 37.2, 53.9, 63.0, 64.7, 78.1, 126.9, 127.5, 129.1, 137.7, 155.3, 156.7, 173.8; ESI MS: *m/z* 353.2 $[M+H]^+$. Anal. calcd. for $C_{17}H_{25}N_3O_5$: C, 58.11; H, 7.17; N, 11.96. Found: C, 58.38; H, 7.52; N, 11.78%.

Boc-Phe-ψ(NH-CO-NH)-Ala-OMe **(5d)**

Yield: 76%; mp 138–139°C; ^{1}H NMR (DMSO): δ 1.15 (3H, d), 1.35 (9H, s), 2.84 (2H, d), 3.65 (3H, s), 3.85 (2H, m), 5.15 (1H, d), 6.35 (1H, d), 6.5 (1H, d), 7.15–7.4 (5H, m); ^{13}C NMR (DMSO): δ 17.1, 28.7, 37.1, 48.7, 54.7, 63.5, 78.3, 126.7, 127.9, 129.2, 137.9, 154.8, 156.3, 174.5; ESI MS: m/z 366.1 $[M+H]^+$. Anal. calcd. for $C_{18}H_{27}N_3O_5$: C, 59.16; H, 7.45; N, 11.50. Found: C, 59.30; H, 7.52; N, 11.20%.

Boc-Ser(Bzl)-ψ(NH-CO-NH)-Gly-OMe **(5e)**

Yield: 78%; mp 124–126°C; ^{1}H NMR (DMSO): δ 1.35 (9H, s), 3.6 (3H, s), 3.7–3.85 (5H, m), 4.48 (2H, s), 5.25 (1H, br), 6.3 (1H, d), 6.45 (1H, d), 7.3–7.4 (5H, m); ^{13}C NMR (DMSO): δ 28.5, 51.7, 57.7, 63.7, 65.0, 71.9, 78.0, 126.6, 129.3, 130.5, 138.0, 155.3, 156.8, 174.5; ESI MS: m/z 383.1 $[M+H]^+$. Anal. calcd. for $C_{18}H_{27}N_3O_6$: C, 56.68; H, 7.15; N, 11.02. Found: C, 56.38; H, 7.20; N, 11.10%.

Boc-Asp(OBzl)-ψ(NH-CO-NH)-Gly-OMe **(5f)**

Yield: 70%; mp 120–121°C; ^{1}H NMR (DMSO): δ 1.4 (9H, s), 2.55 (2H, d), 3.6 (3H, s), 3.85 (3H, m), 5.0 (1H, m), 5.1 (2H, s), 6.35 (1H, d), 6.5 (1H, t), 7.25–7.35 (5H, m); ^{13}C NMR (DMSO): δ 28.1, 38.0, 49.5, 61.5, 62.7, 64.7, 78.5, 127.6, 128.5, 129.3, 136.7, 155.0, 156.8, 174.5; ESI MS: m/z 425.1 $[M+H]^+$. Anal. calcd. for $C_{20}H_{29}N_3O_7$: C, 56.73; H, 6.90; N, 9.92. Found: C, 56.80; H, 6.52; N, 10.38%.

Boc-Glu(OBzl)-ψ(NH-CO-NH)-Leu-OMe **(5g)**

Yield: 74%; mp 139–140°C; ^{1}H NMR (DMSO): δ 0.91 (6H, d), 1.3–1.45 (11H, s), 1.65 (1H, m), 2.55 (2H, m), 2.9 (2H, m), 3.65 (3H, s), 3.8–3.9 (2H, m), 5.15 (2H, s), 5.3 (1H, d), 6.35 (1H, d), 6.5 (1H, d), 7.3–7.4 (5H, m); ^{13}C NMR (DMSO): δ 22.1, 23.1, 24.7, 28.6, 37.9, 39.7, 41.9, 50.9, 51.7, 61.9, 63.1, 78.7, 126.7, 127.6, 128.9, 137.7, 155.3, 156.8, 157.5, 178.1; ESI MS: m/z 480.2 $[M+H]^+$. Anal. calcd. for $C_{24}H_{37}N_3O_7$: C, 60.11; H, 7.78; N, 8.76. Found: C, 59.80; H, 7.52; N, 9.10%.

Boc-Ile-ψ(NH-CO-NH)-Phe-OMe **(5h)**

Yield: 72%; mp 135–137°C; ^{1}H NMR (DMSO): δ 0.91 (6H, m), 1.13 (1H, m), 1.35 (9H, s), 1.55 (2H, m), 2.85 (2H, d), 3.65 (3H, s), 3.85 (2H, m), 5.15 (1H, d),

6.35 (1H, d), 6.5 (1H, d), 7.3–7.45 (5H, m); ^{13}C NMR (DMSO): δ 11.7, 15.5, 25.5, 28.3, 37.5, 55.1, 58.7, 63.5, 78.3, 126.7, 127.9, 129.2, 137.9, 154.8, 156.3, 174.5; ESI MS: m/z 409.4 $[M+H]^+$. Anal. calcd. for $C_{21}H_{33}N_3O_5$: C, 61.90; H, 8.16; N, 10.31. Found: C, 62.20; H, 8.52; N, 10.38%.

Boc-Tyr-ψ(NH-CO-NH)-Ala-OMe **(5i)**

Yield: 70%; mp 140–42°C; ^{1}H NMR (DMSO): δ 1.2 (3H, s), 1.37 (9H, s), 3.1 (2H, m), 3.71 (3H, s), 3.8 (m, 1H), 6.2 (m, 2H), 7.0 (m, 4H); ^{13}C NMR (DMSO): δ 16.1, 28.0, 40.3, 50.6, 52.1, 66.9, 81.0, 119.1, 127.1, 128.7, 128.4, 1548, 158.4, 159.6, 172.1; ESI MS: m/z 382.4 $[M+H]^+$. Anal. calcd. for $C_{18}H_{27}N_3O_8$: C, 56.68; H, 7.13; N, 11.02; O, 25.17. Found: C, 56.62; H, 7.18; N, 11.06, O, 25.20%.

Boc-Trp(Z)-ψ(NH-CO-NH)-Val-OMe **(5j)**

Yield: 69%; mp 148–50°C; ^{1}H NMR (DMSO): δ 0.9 (6H, s), 1.36 (9H, s), 1.8 (m, 1H), 3.4 (m, 2H), 3.7 (3H, s), 4.4 (1H, m), 5.2 (2H, s), 5.8 (m, 1H), 7.0–7.9 (10H, m); ^{13}C NMR (DMSO): δ 18.1, 28.0, 34.2, 34.3, 57.2, 29.8, 63.4, 67.7, 79.3, 108.8, 119.2, 122.2, 122.5, 123.7, 124.8, 127.6, 127.9, 128.5, 136.0, 145.4, 146.8, 159.8, 161.0, 173.1; ESI MS: m/z 567.3 $[M+H]^+$. Anal. calcd. for $C_{30}H_{38}N_4O_7$: C, 63.59; H, 6.76; N, 9.89; O, 19.76. Found: C, 63.55; H, 6.80; N, 9.91; O, 19.80%.

Boc-Dipeptidyl Urea Acids (8a–d): General Procedure

To a stirred suspension of amino acid **6** (1 mmol) in CH_2Cl_2 (5 mL) freshly distilled TMS-Cl (2.2 mmol) and TEA (2.2 mmol) were added and refluxed for 2 h. The reaction mixture was cooled to rt, *O*-succinimidyl methylcarbamate **3** (1 mmol) in THF–DMF (2:1, 5 mL) solution was added, and it was stirred for 30 min at rt. After the completion of the reaction, the solvent was evaporated under reduced pressure, and the residue was triturated with water (to remove side product formed during the reaction), filtered, and recrystallized using DMSO–water to afford the pure urea acids **8a–d** as crystalline off-white solids.

Data

Boc-Leu-ψ(NH-CO-NH)-Ala-OH **(8a)**

Yield: 80%; mp 140–141°C; ^{1}H NMR (DMSO): δ 0.93 (6H, d), 1.15 (3H, d), 1.3 (9H, s), 1.35 (2H, t), 1.6 (1H, m), 3.75–3.8 (2H, m), 5.2 (1H, br),

6.3–6.45 (2H, m), 8.4 (1H, d); ^{13}C NMR (DMSO): δ 17.4, 22.1, 23.3, 24.7, 28.5, 40.1, 49.2, 51.3, 78.1, 155.2, 156.9, 177.8; MS (FAB$^+$): m/z 318.1. Anal. calcd. for $C_{14}H_{27}N_3O_5$: C, 52.98; H, 8.57; N, 13.24. Found: C, 52.66; H, 8.57; N, 13.20%.

Boc-Phe-ψ(NH-CO-NH)-Ala-OH **(8b)**

Yield: 78%; mp 137–138°C; ^{1}H NMR (DMSO): δ 1.16 (3H, d), 1.31 (9H, s), 2.85 (2H, d), 3.8–3.9 (2H, m), 5.3 (1H, br), 6.4–6.4 (2H, m), 7.15–7.3 (5H, m), 8.35 (1H, d); ^{13}C NMR (DMSO):δ 17.3, 28.1, 37.5, 48.7, 54.1, 77.2, 126.7, 129.1, 130.2, 137.9, 155.1, 156.7, 177.5; MS (FAB$^+$): m/z 352.2. Anal. calcd. for $C_{17}H_{25}N_3O_5$: C, 58.11; H, 7.17; N, 11.96. Found: C, 58.16; H, 7.40; N, 11.70%.

Boc-Leu-ψ(NH-CO-NH)-Gly-OH **(8c)**

Yield: 75%; mp 123–125°C; ^{1}H NMR (DMSO): δ 0.93 (6H, d), 1.3 (9H, s), 1.4 (2H, m), 1.65 (1H, m), 3.5 (2H, d), 3.7 (1H, m), 5.2 (1H, d), 6.1 (1H, br), 6.35 (1H, d), 8.35 (1H, d); ^{13}C NMR (DMSO): δ 22.0, 23.1, 24.7, 28.1, 40.5, 41.9, 52.0, 75.9, 155.2, 156.9, 178.8; MS (FAB$^+$): m/z 304.2. Anal. calcd. for $C_{13}H_{25}N_3O_5$: C, 51.47; H, 8.31; N, 13.85. Found: C, 51.20; H, 8.52; N, 13.78%.

Boc-Val-ψ(NH-CO-NH)-Phe-OH **(8d)**

Yield: 72%; mp 128–130°C; ^{1}H NMR (DMSO): δ 0.80–0.91 (6H, t), 1.35 (9H, s), 1.85 (1H, m), 2.8 (2H, d), 4.3 (1H, m), 4.8 (1H, m), 5.5 (1H, d), 6.1–6.3 (2H, br), 7.13–7.25 (5H, m), 8.35 (1H, d); ^{13}C NMR (DMSO): δ 18.5, 19.3, 26.1, 29.2, 37.2, 54.9, 62.1, 78.1, 126.7, 128.5, 129.2, 137.5, 154.5, 156.7, 176.2; MS (FAB$^+$): m/z 380.0. Anal. calcd. for $C_{19}H_{29}N_3O_5$: C, 60.14; H, 7.70; N, 11.07. Found: C, 60.45; H, 7.57; N, 11.20%.

Test for Racemization of Methylcarbamate 3f

To a stirred solution of methylcarbamate **3f** (0.377 g, 1 mmol) in THF-DMF (5 mL), optically pure (R)-(+)-1-phenylethylamine (0.121 g, 1 mmol) was added, and the mixture was stirred at 25°C for 30 min. After routine workup, it resulted in the diastereomer **10a**. The solid was recrystallized using DMSO–water. Mp 165°C; $[\alpha]_D^{25}$: +24.8 (c 1, DMSO); ^{1}H NMR (DMSO): δ 1.2–1.4 (12H, t), 2.9 (2H, d), 3.3 (1H, m), 4.7 (1H, m),

5.1 (1H, br), 6.2 (1H, s), 6.55 (1H, m), 7.2–7.35 (10H, m); MS (FAB^+): m/z 384.1. When this experiment was repeated using **3f** with optically pure (S)-(−)-1-phenylethylamine (0.121 g, 1 mmol), the other diastereomer **10b** was isolated. Mp 160–161°C; $[\alpha]_D^{25}$: −25.6 (c 1, DMSO); ^{1}H NMR (DMSO): δ 1.2–1.4 (12H, t), 2.9 (2H, d), 3.3 (1H, m), 4.7 (1H, m), 5.1 (1H, br), 6.2 (1H, s), 6.55 (1H, m), 7.2–7.35 (10H, m); MS (FAB^+): m/z 384.1. Anal. calcd. for $C_{22}H_{29}N_3O_3$: C, 68.90; H, 7.62; N, 10.96. Found: C, 68.91; H, 7.23; N, 10.59%. The experiment was repeated using **3f** with racemic 1-phenylethylamine (0.121 g, 1 mmol). The corresponding diastereomer **10c** was obtained and isolated. Mp 155–157°C; ^{1}H NMR (DMSO): δ 1.2–1.4 (12H, dd), 2.9 (2H, d), 3.3 (1H, m), 4.7 (1H, m), 5.1 (1H, br), 6.2 (1H, s), 6.55 (1H, m), 7.2–7.35 (10H, m); MS (FAB^+): m/z 384.0. Anal. calcd. for $C_{22}H_{29}N_3O_3$: C, 68.90; H, 7.62; N, 10.96. Found: C, 68.81; H, 7.29; N, 10.60%.

ACKNOWLEDGMENTS

Authors thank the Department of Science and Technology, Government of India, for financial assistance. Authors also thank Sophisticated Instruments Facility, Indian Institute of Science (IISc), for NMR data and P. Balaram, Molecular Biophysics Unit (MBU), IISc, Bangalore, for mass spectral data.

REFERENCES

1. Zhang, X.; Rodrigues, J.; Evans, L.; Hinkle, B.; Ballantyne, L.; Pena, M. Formation of urea dipeptides from carbonyldiimidazole: Application toward the protease inhibitors GE 20372 and MAPI. *J. Org. Chem.* **1997**, *62*, 6420–6423.
2. Tamilarasu, N.; Huq, I.; Rana, T. M. Targeting RNA with peptidomimetic oligomers in human cells. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 505–507.
3. Holmes, D. L.; Smith, E. M.; Nowick, J. S. Solid-phase synthesis of artificial ?-sheets. *J. Am. Chem. Soc.* **1997**, *119*, 7665–7669.
4. Nowick, J. S.; Pairish, M.; Lee, I. Q.; Holmes, D. L.; Ziller, J. W. An extended ?-strand mimic for a larger artificial ?-sheet. *J. Am. Chem. Soc.* **1997**, *119*, 5413–5424
5. Fischer, V.; Semetey, V.; Lozano, J-M.; Schaffner, A-P.; Briand, J-P.; Didierjean, C.; Guichard, G. Succinimidyl carbamate derivatives from N-protected ?-amino acids and dipeptides- synthesis of ureidopeptides and oligourea/peptide eptide hybrids. *Eur. J. Chem.* **2007**, 2511–2525, and references therein.
6. Okada, Y.; Tsuda, Y.; Yagyu, M. Amino acids and peptides. VI. Curtius rearrangement of acyl amino acid and peptide azides and reactivity of the isocyanates. *Chem. Pharm. Bull.* **1980**, *28*, 2254.

7. Guichard, G.; Semetey, V.; Didierjean, C.; Aubry, A.; Briand, J.-P.; Rodriguez, M. Effective preparation of O-succinimidyl-2-(tert-butoxycarbonylamino)ethylcarbamate derivatives from ?-amino acids. Application to the synthesis of urea-containing pseudopeptides and oligoureas. *J. Org. Chem.* **1999**, *64*, 8702.
8. Guichaard, G.; Semetey, V.; Rodriguez, M.; Briand, J.-P. Solid phase synthesis of oligoureas using O-succinimidyl-(9H-fluoren-9-ylmethoxycarbonylamino)ethylcarbamate derivatives as activated monomers. *Tetrahedron Lett.* **2000**, *41*, 1553.
9. Semetey, V.; Hemmerlin, C.; Didierjean, C.; Schaffner, A.-P.; Giner, A. G.; Aubry, A.; Briand, J.-P.; Marraud, M.; Guichard, G. Unexpected stability of the urea cistrans isomer in urea-containing model pseudopeptides. *Org. Lett.* **2001**, *3*, 3843.
10. Semetey, V.; Schaffner, A.-P.; Marraud, M.; Didierjean, C.; Aubry, A.; Rodriguez, M.; Briand, J.-P.; Guichard, G. In *Peptides*; J. Martinez, J.-A. Fehrentz (Eds.), The 26th European Peptide Symposium, Montpellier, EDK, Paris, 2000, pp. 161–162.