# Alternative and Chemoselective Deprotection of the α-Amino and Carboxy Functions of *N*-Fmoc-Amino Acid and *N*-Fmoc-Dipeptide Methyl Esters by Modulation of the Molar Ratio in the $AlCl_3$/*N*,*N*-Dimethylaniline Reagent System

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The amino and carboxy functions in *N*-Fmoc-α-amino acid and *N*-Fmoc-peptide methyl esters can be alternatively and chemoselectively deprotected by treatment with the reagent system $AlCl_3$/*N*,*N*-dimethylaniline (DMA). The chemoselectivity of the process is controlled by modulating the relative molar ratio of the Lewis acid and DMA.


## Introduction

The use of orthogonal protecting groups to mask the α-amino, side chain and terminal carboxy functions is of great importance for successful peptide synthesis, both in solution and under solid phase conditions.

The protection of the terminal carboxy moiety is a very important step in solution peptide synthesis. From previous experience, in many applications it would be extremely convenient to mask the carboxy group as a methyl ester[1,2] when the 9-fluorenylmethyloxycarbonyl group is used to protect the terminal α-amino function.

The availability of simple and straightforward procedures that would allow the alternative unblocking of the carboxy or α-amino functions with high chemoselectivity represents a challenging field in the synthesis of peptides, especially when peptide chains are synthesized by a building-block approach.[3,4] Alternative deprotection of amino and carboxy functions could also be useful in multi-step total synthesis.

## Results and Discussion

The selective cleavage of an ester function without removing the Fmoc protecting group on the amino moiety[5−21] is an important strategic element in peptide synthesis. In a previous paper,[22] we reported a facile procedure for the chemoselective unblocking of the carboxy group in *N*-Fmoc-protected α-amino acid and peptide methyl esters using the reagent system $AlCl_3$/*N*,*N*-dimethylaniline (DMA). We also reported[23,24] that the reagent system $AlCl_3$/toluene easily removes the *N*-9-fluorenylmethyloxycarbonyl protecting group from the amino function of α-amino acid and short peptide methyl esters without demolition of the methyl ester moiety and with retention of the configuration of the carbon atoms of the peptide systems.

The analogy between the two reagent systems containing the same Lewis acid species, $AlCl_3$, which remove either the ester function ($AlCl_3$/DMA) or the urethane group ($AlCl_3$/toluene), cannot provide a clear explanation for the diverse behaviour observed for the two reagent systems considered (Scheme 1).

We have carried out some experiments in order to establish the optimal molar ratio of the constituents in the reagent system $AlCl_3$/DMA for removal of the Fmoc group from the α-nitrogen atom. Fast chemoselective removal of the Fmoc protecting group was obtained with a 10:4 molar ratio of $AlCl_3$ and DMA (Scheme 2, Table 1).

The compatibility of the reagent system $AlCl_3$/DMA with aromatic moieties on the side chains of *N*-Fmoc-amino acid methyl esters, was checked by treating *N*-Fmoc-phenylalanine methyl ester (**1b**) and *N*-Fmoc-tyrosine methyl ester (**1d**) under the conditions typically used for the chemoselective removal of the Fmoc group (molar ratio $AlCl_3$/DMA. 10:4). The *N*-deprotected methyl esters obtained were converted, by treatment with acetic anhydride, into the corresponding *N*-acetyl derivatives **2b** and **2d** (Scheme 2) in 77% and 94% yields, respectively. Successively, the same substrates **1b** and **1d** were treated with $AlCl_3$/DMA in a 7:11 molar ratio to unblock the carboxy function. Under these conditions, the reagent system $AlCl_3$/DMA operates by removing the methyl group from the ester function of **1b** and

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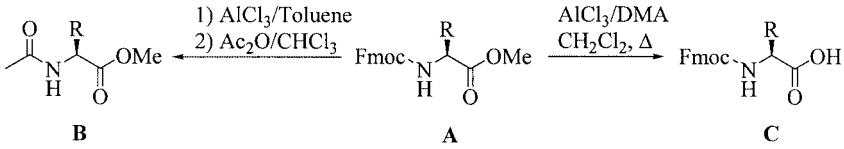
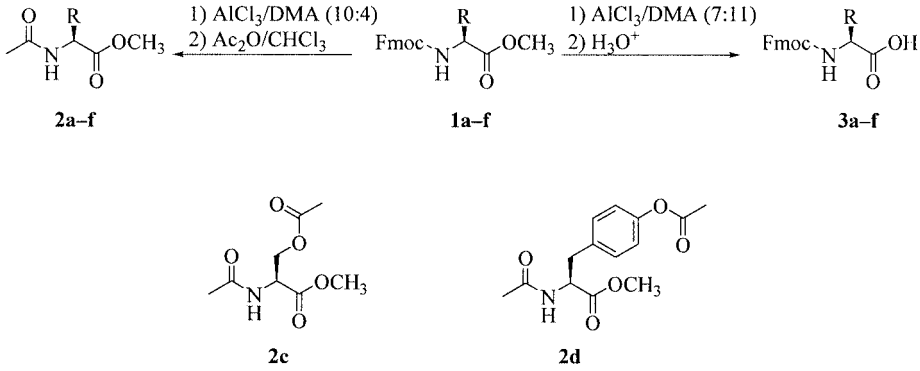



Scheme 1




Scheme 2

Table 1. Yields of compounds **2a**−**f** and **3a**−**f**

| | R | Yield (%) **2** | **3** |
|---|---|---|---|
| **a** | $CH(CH_3)CH_2CH_3$ | 83 | 84 |
| **b** | $CH_2Ph$ | 77 | 84 |
| **c** | $CH_2OH$ | 93 | 83 |
| **d** | $CH_2C_6H_4OH$ | 94 | 78 |
| **e** | $CH_2CH_2SCH_3$ | 98 | 98 |
| **f** | $CH_2SCH_2NHCOCH_3$ | 67 | 83 |

**1d** without demolition of the urethane moiety present on the α-nitrogen atom. In this case, the treatment afforded *N*-Fmoc-phenylalanine (**3b**) and *N*-Fmoc-tyrosine (**3d**) in 84% and 78% yields, respectively. In all the reactions performed with the substrates **1b** and **1d**, either the removal of the Fmoc protecting group or, alternatively, the demethylation of the ester function, were chemoselective as determined by chromatographic analysis of the respective reaction mixtures. Furthermore, the experimental data confirmed that the reagent system $AlCl_3$/DMA does not promote the formation of side products arising from possible electrophilic attacks of the methyl and 9-fluorenylmethyl cations on the aromatic rings of the side chains present in the starting protected substrates.

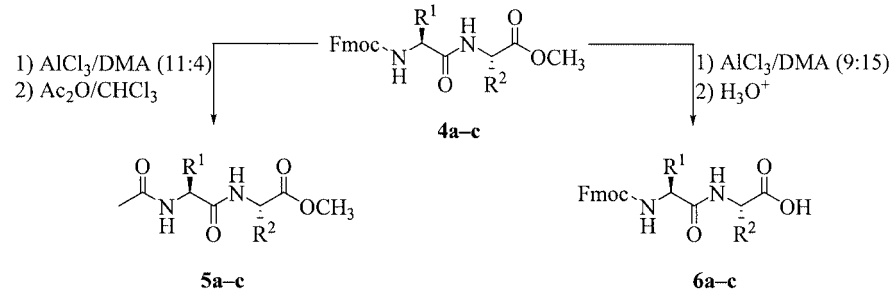
In order to establish the capabilities of the reagent system $AlCl_3$/DMA, other *N*-Fmoc-α-amino acid methyl esters, characterized by a nucleophilic site on the side chains, were subjected to both the reaction conditions described above. This new set of experiments allowed the evaluation of the possible competition between the reactions of the nucleophilic sites and DMA with the 9-fluorenylmethyl or, alternatively, the methyl cations generated during the removal of the Fmoc protecting group and the demethylation of the carboxymethyl function, respectively.

Substrates **1c** and **1e** were treated under the appropriate experimental conditions, which regulate each of the two different reactive pathways, to allow the specific unblocking of the amino and the carboxy functions. The results obtained are summarized in Table 1. Upon treatment with the reagent system $AlCl_3$/DMA in the 7:11 molar ratio, the two substrates generated the corresponding *N*-Fmoc-amino acids **3c** and **3e** in 83% and 98% yields, respectively. When a 10:4 molar ratio of the Lewis acid and DMA was applied instead, the corresponding methyl esters were recovered and isolated as the respective *N*-acetyl derivatives **2c** and **2e** in 93% and 98% yields, respectively, after treatment with $Ac_2O$. Mass spectrometric and $^1H$ NMR analysis of the crude materials resulting from these reactions revealed that alkylation of the nucleophilic functionalities on the side chains of the starting substrates does not occur under the experimental conditions used. The *N*-Fmoc-*S*-(acetamidomethyl)cysteine methyl ester (**1f**) was also subjected to both experimental conditions. Compounds **2f** and **3f**, obtained by removing the urethanic or the methyl ester function, respectively, preserve the acetamidomethyl protecting group on the side chain.

The chemoselectivity of the removal of the Fmoc protecting group or, alternatively, the demethylation of the ester function was also studied in simple peptide systems. Dipeptides **4a**−**c** (Scheme 3) were treated with the reagent system $AlCl_3$/DMA in the molar ratio 11:4. This new composition of the reagent system used for deprotection was justified by the higher number of functionalities present in the dipeptides that are capable of coordinating to the electrophilic reagent.

The intermediate dipeptides, which bear a free terminal amino function, were quenched by acetylation with $Ac_2O$. The *N*-acetylated dipeptidyl derivatives **5a**−**c** were recovered in yields ranging from 71% to 74% (Table 2). The

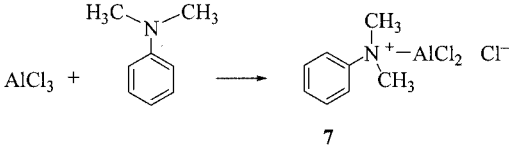



Scheme 3

Table 2. Yields of compounds **5a**−**c** and **6a**−**c**

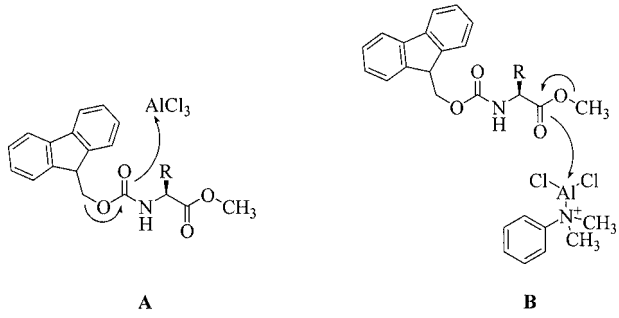
| | $R^1$ | $R^2$ | Yield (%) **5** | **6** |
|---|---|---|---|---|
| **a** | $CH_2CH(CH_3)_2$ | $CH_2Ph$ | 71 | 78 |
| **b** | $CH(CH_3)_2$ | $CH_2CH(CH_3)_2$ | 73 | 85 |
| **c** | $CH_2Ph$ | $CH(CH_3)_2$ | 74 | 75 |

same substrates **4a**−**c**, upon treatment with the reagent system $AlCl_3$/DMA in a molar ratio of 9:15, gave the *N*-Fmoc protected dipeptides **6a**−**c** (Scheme 3) in 75−85% yields (Table 2). Two different reaction pathways generate the peptide systems; the carboxymethyl moiety is maintained whilst the urethane protecting group is removed or, alternatively, the protection on the α-nitrogen atom is retained and the methyl from the ester function is lost. This particular behaviour could be of great importance when the availability of differently protected dipeptide precursors is required for the preparation of more complex peptides through a building-block synthetic strategy. The different compositions of the reagent system $AlCl_3$/DMA can satisfactory explain both the reaction pathways observed for the alternative and chemoselective removal of the protecting groups in *N*-Fmoc-α-amino acid and *N*-Fmoc-peptide methyl esters. The species responsible for the unblocking are different in the two cases studied. The 1:1 $AlCl_3$/DMA adduct **7** (Scheme 4) plays a key role in the deprotection of the substrates analysed in this study.




Scheme 4

The formation of the equimolecular adduct of the Lewis acid and DMA could explain the dissimilarities observed when different molar ratios of $AlCl_3$ and DMA are used to compose the reagent system. Both the adduct **7** and $AlCl_3$ are the real species present in the reaction mixtures when the reagent system has an excess of $AlCl_3$. The free Lewis acid coordinates to the most basic site, the carbonyl oxygen




Scheme 5

atom of the urethane protecting group (**A**, Scheme 5). This moiety, upon reaction with $AlCl_3$, generates the transient 9-fluorenylmethyl cation, which is quenched by the adaptable adduct **7**, inducing the demolition of the masking group placed on the α-nitrogen atom. The adduct between DMA and the 9-fluorenylmethyl cation was identified by GC-MS analysis of the crude material recovered after work up of the reaction mixtures.

When the reagent system is prepared with an excess of DMA, the reactive Lewis acid is **7** (**B**, Scheme 5), which is more sterically hindered and cannot coordinate to the urethane carbonyl group and thus prefers to attack the ester function. The consequent demethylation proceeds via formation of the methyl cation, which in turn is quenched by DMA present in good excess with respect to the Lewis acid. In this case the adduct produced from the reaction between DMA and the methyl cation was also identified by GC-MS analysis. Alternatively it can be supposed that the DMA demethylates, by a $S_N2$ mechanism, the ester function activated by the $AlCl_3$/DMA adduct because of the absence in the crude reaction materials of methylated products in the side chains.

## Conclusion

The chemoselective removal of the Fmoc protecting group from the α-amino function or, alternatively, demethylation of the ester moiety in *N*-Fmoc-α-amino acid and peptide methyl esters can be performed by modulating the molar ratio of the two components of the reagent system.

Changes in the molar ratio of $AlCl_3$ and DMA correspond to changes in the reactive species responsible for the removal of the target protecting group.

## Experimental Section

**General Remarks:** All reagents were purchased from Aldrich Co. and Senn Chemical. All solvents were purified and dried by standard procedures and distilled prior to use. Melting points were determined with a Kofler hot-stage apparatus and are uncorrected. NMR characterization of all the compounds was performed with a Bruker Avance 300 spectrometer. $^1H$ and $^{13}C$ NMR spectra were recorded at 300 MHz and 75.5 MHz, respectively, with $CDCl_3$ or [$D_6$]DMSO as solvent. GC-MS analyses were carried out using a 30-m HP-35MS capillary column with a 0.25 mm internal diameter and a 0.25 μm film thickness. The mass detector was operated in the electron impact ionization mode (EI-MS) at an electron energy of 70 eV. Mass spectra were recorded with a Vacuum Generator ZAB-2F spectrometer, with 3-nitrobenzyl alcohol as the matrix, by fast atom bombardment ($FAB^+$ MS) with a neutral Xenon beam operating at 8 keV and a total current of 10 μA. Reaction mixtures were monitored by TLC with silica gel 60-F254 precoated glass plates purchased from Merck. Short column flash chromatography (SCFC) was performed on Kieselgel 60 H without gypsum. All reactions were carried out under an inert atmosphere ($N_2$).

**Removal of the Fmoc Group of *N*-Fmoc-α-Amino Acid Methyl Esters 1a−f. General Procedure:** $AlCl_3$ (10 mmol) and *N*,*N*-dimethylaniline (4 mmol) were added to a suspension of the appropriate *N*-Fmoc-α-amino acid methyl ester **1a−f** (1 mmol) in dry dichloromethane (10 mL). The resulting mixture was stirred under reflux for 4 h until TLC analysis of the reaction mixture (chloroform/methanol, 90:10, v/v) showed complete conversion of the precursor. Methanol was then added dropwise until effervescence stopped, and the solvents were evaporated to dryness. The chromatographic purification of the crude product (chloroform/methanol, 95:5, v/v) afforded the corresponding α-amino acid methyl ester, which was successively dissolved in an aqueous 5% $Na_2CO_3$ solution (5 mL) and treated with acetic anhydride (3 mL) in dichloromethane (10 mL). After 2 h at room temp., the organic layer was separated and the aqueous layer was extracted with dichloromethane (3 × 3 mL). The combined organic extracts were washed once with brine, dried with $Na_2SO_4$ and then evaporated to dryness to give the *N*-acetyl-α-amino esters **2a−f** in 67−98% overall yields. The compound characterization data for **2a−f** fitted those observed for authentic samples of *N*-acetyl-α-protected amino esters.

**Removal of the Methyl Ester Group of *N*-Fmoc-α-Amino Acid Methyl Esters 1a−f:**[22] A suspension of the appropriate *N*-Fmoc-α-amino acid methyl ester **1a−f** (1 mmol) in dry dichloromethane was treated with the reagent system $AlCl_3$ (7 mmol)/*N*,*N*-dimethylaniline (11 mmol). Chromatographic purification of the crude reaction products enabled a simple removal of the excesses of the reagents used to afford the respective *N*-Fmoc-amino acids **3a−f** in high yields. The compound characterization data for **3a−f** fitted those observed for authentic samples of *N*-Fmoc-protected amino acids.

**Synthesis of Dipeptides 4a−c:** A solution of the appropriate *N*-Fmoc-α-amino acid chloride (0.9 mmol) in ethanol-free chloroform (10 mL) was added dropwise to a magnetically stirred solution of the commercially available α-amino ester hydrochlorides (1 mmol) dissolved in an aqueous 5% $Na_2CO_3$ solution (5 mL). The resulting mixture was stirred at room temp. for 2 h. The organic layer was separated and the aqueous layer was extracted with chloroform (2 × 5 mL). The combined organic extracts were washed with aqueous 1 M HCl (2 × 3 mL), washed once with brine, dried with $Na_2SO_4$ and then evaporated to dryness to give the dipeptides **4a−c** in 86−92% overall yields.

The compound characterization data for **4b** fitted those observed for an authentic sample of *N*-Fmoc-valyl-leucine methyl ester.[22]

**Compound 4a:** White solid (1.45 g, 86% yield), m.p. 137−140 °C. IR (KBr): $\tilde{\nu}$ = 3305, 3090, 2965, 1742, 1692, 1641, 1532, 1241, 760, 741 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): δ = 0.88−0.98 [m, 6 H, $CH_2CH(CH_3)_2$], 1.45−1.70 [m, 3 H, $CH_2CH(CH_3)_2$], 2.98 (dd, $J_1$ = 5.8, $J_2$ = 13.9 Hz, 1 H, $CH_2Ph$), 3.13 (dd, $J_1$ = 5.9, $J_2$ = 13.9 Hz, 1 H, $CH_2Ph$), 3.72 (s, 3 H, $OCH_3$), 4.20 (m, 1 H, Fmoc-CH), 4.30−4.51 (m, 3 H, CHNH), 4.85 (m, 1 H, $CHCH_2Ph$), 5.21 (d, $J$ = 8.4 Hz, 1 H, OCONH), 6.51 (d, $J$ = 7.4 Hz, 1 H, CHCONH), 7.01−7.28 (m, 5 H, Ar-H), 7.31−7.83 (m, 8 H, Ar-H) ppm. $^{13}C$ NMR: δ = 15.3, 22.0, 22.9, 24.6, 37.8, 41.4, 47.1, 53.2, 53.4, 67.1, 120.0, 125.0, 125.1, 127.1, 127.2, 127.7, 128.6, 129.3, 136.6, 141.3, 143.8, 171.7, 171.8 ppm. $FAB^+$ MS: *m*/*z* (%) = 515 (12) $[M + H]^+$, 179 (100), 178 (78), 165 (39). $C_{31}H_{34}N_2O_5$ (514.61): calcd. C 72.35, H 6.66, N 5.44, O 15.55; found C 72.45, H 6.63, N 5.40.

**Compound 4c:** White solid (1.62 g, 91% yield), m.p. 167−169 °C. IR (KBr): $\tilde{\nu}$ = 3300, 3078, 2964, 1744, 1701, 1654, 1542, 1261, 758, 739 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): δ = 0.81 [d, $J$ = 6.8 Hz, 3 H, $CH(CH_3)_2$], 0.87 [d, $J$ = 6.9 Hz, 3 H, $CH(CH_3)_2$], 2.10 [m, 1 H, $CH(CH_3)_2$], 3.16−3.25 (m, 2 H, $CH_2Ph$), 3.71 (s, 3 H, $OCH_3$), 4.20 (m, 1 H, $CHCH_2Ph$), 4.30−4.55 (m, 4 H, Fmoc-$CH_2$, $CHCH(CH_3)_2$, Fmoc-CH), 5.53 (d, $J$ = 7.1 Hz, 1 H, OCONH), 6.41 (d, $J$ = 7.5 Hz, 1 H, CHCONH), 7.15−7.88 (m, 13 H, Ar-H) ppm. $^{13}C$ NMR: δ = 17.6, 30.5, 37.5, 47.1, 51.9, 55.1, 55.7, 67.4, 126.0, 126.8, 127.8, 128.2, 128.4, 128.8, 129.0, 139.5, 141.0, 143.6, 156.0, 171.6, 171.8 ppm. $FAB^+$ MS: *m*/*z* (%) = 501 (100) $[M + H]^+$, 279 (61), 179 (62), 178 (38), 165 (25). $C_{30}H_{32}N_2O_5$ (500.59): calcd. C 71.98, H 6.44, N 5.60, O 15.98; found C 72.09, H 6.40, N 5.57.

**Removal of the Fmoc Group of Dipeptides 4a−c. General Procedure:** $AlCl_3$ (11 mmol) and *N*,*N*-dimethylaniline (4 mmol) were added to a suspension of the appropriate dipeptide **4a−c** (1 mmol) in dry dichloromethane (10 mL). The resulting mixture was stirred under reflux for 4 h, until TLC analysis (chloroform/methanol, 90:10, v/v) showed complete conversion of the precursor. Methanol was then added dropwise until effervescence stopped and the solvents were evaporated to dryness. The chromatographic purification of the crude product (chloroform/methanol, 95:5, v/v) afforded the corresponding dipeptides which were successively dissolved in an aqueous 5% solution of $Na_2CO_3$ (5 mL) and treated with acetic anhydride (3 mL) in chloroform (10 mL). After 2 h at room temp., the organic layer was separated and the aqueous layer was extracted with dichloromethane (3 × 3 mL). The combined organic extracts were washed once with brine, dried with $Na_2SO_4$ and then evaporated to dryness to give the dipeptides **5a−c** in 71−74% overall yields.

**Compound 5a:** Oil (0.37 g, 71% yield). IR (neat): $\tilde{\nu}$ = 3275, 3048, 2968, 1749, 1657, 1642, 1251 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): δ = 0.90−0.95 [m, 6 H, $CH_2CH(CH_3)_2$], 1.53−1.70 [m, 3 H, $CH_2CH(CH_3)_2$, $CH_2CH(CH_3)_2$], 1.99 (s, 3 H, $CH_3CONH$), 3.09 (dd, $J_1$ = 6.5, $J_2$ = 13.7 Hz, 1 H, $CH_2Ph$), 3.18 (dd, $J_1$ = 5.5, $J_2$ = 13.7 Hz, 1 H, $CH_2Ph$), 3.74 (s, 3 H, $OCH_3$), 4.45 [m, 1 H, $CHCH_2CH(CH_3)_2$], 4.87 (m, 1 H, $CHCH_2Ph$), 5.82 (d, $J$ = 8.2 Hz, 1 H, OCONH), 6.51 (d, $J$ = 7.5 Hz, 1 H, CHCONH), 7.09−7.30

(m, 5 H, Ar-*H*) ppm. $^{13}$C NMR: δ = 22.1, 22.6, 22.9, 37.0, 41.2, 51.1, 51.9, 53.2, 126.0, 127.8, 128.7, 139.5, 170.7, 171.6, 171.8 ppm. EI-MS: *m*/*z* (%) = 334 (2) [M$^{+}$·], 162 (40), 128 (74), 86 (100), 44 (17), 43 (14). $C_{18}H_{26}N_2O_4$ (334.41): calcd. C 64.65, H 7.84, N 8.38, O 19.14; found C 64.75, H 7.81, N 8.34.

**Compound 5b:** Yellow solid (0.40 g, 73% yield), m.p. 112−114 °C. IR (KBr): ṽ = 3278, 2963, 1744, 1672, 1643, 1264 cm$^{-1}$. $^1$H NMR ($CDCl_3$): δ = 0.91 [d, *J* = 2.8 Hz, 3 H, $CH_2CH(CH_3)_2$], 0.93 [d, *J* = 2.9 Hz, 3 H, $CH_2CH(CH_3)_2$], 0.96 [d, *J* = 6.6 Hz, 3 H, $CH(CH_3)_2$], 0.99 [d, *J* = 6.5 Hz, 3 H, $CH(CH_3)_2$], 1.50−1.70 [m, 3 H, $CH_2CH(CH_3)_2$], 2.01 (s, 3 H, $CH_3CONH$), 2.07 [m, 1 H, $CH(CH_3)_2$], 3.73 (s, 3 H, $OCH_3$), 4.40 [dd, $J_1$ = 7.2, $J_2$ = 8.9 Hz, 1 H, $CHCH(CH_3)_2$], 4.55 [m, 1 H,$CHCH_2CH(CH_3)_2$], 6.55 (d, *J* = 8.9 Hz, 1 H, OCONH), 6.85 (d, *J* = 7.7 Hz, 1 H, CHCONH) ppm. $^{13}$C NMR: δ = 18.3, 18.9, 21.8, 22.7, 23.2, 24.7, 31.5, 40.9, 50.9, 52.2, 58.2, 170.1, 171.5, 173.1 ppm. EI-MS: *m*/*z* (%) =286 (1) [M$^{+}$·], 230 (5), 144 (85), 100 (14), 86 (38), 72 (100), 55 (11), 43 (17). $C_{14}H_{26}N_2O_4$ (286.37): calcd. C 58.72, H 9.15, N 9.78, O 22.35; found C 58.82, H 9.11, N 9.74.

**Compound 5c:** Oil (0.53 g, 74% yield). IR (neat): ṽ = 3287, 3055, 2959, 1746, 1658, 1635, 1263 cm$^{-1}$. $^1$H NMR ($CDCl_3$): δ = 0.82 [d, *J* = 6.8 Hz, 3 H, $CH(CH_3)_2$], 0.90 [d, *J* = 6.8 Hz, 3 H, $CH(CH_3)_2$], 2.01 (s, 3 H, $CH_3CONH$), 2.08 [m, 1 H, $CH(CH_3)_2$], 3.00−3.10 (m, 2 H, $CH_2Ph$), 3.70 (s, 3 H, $OCH_3$), 4.45 [dd, $J_1$ = 5.0, $J_2$ = 8.1 Hz, 1 H, $CHCH(CH_3)_2$], 4.70 (m, 1 H, $CHCH_2Ph$), 6.23 (d, *J* = 7.5 Hz, 1 H, OCONH), 6.32 (d, *J* = 8.1 Hz, 1 H, CHCONH), 7.20−7.38 (m, 5 H) ppm. $^{13}$C NMR: δ = 17.7, 18.7, 29.7, 31.1, 38.2, 40.7, 52.2, 54.5, 57.4, 112.7, 116.7, 127.0, 128.7, 170.2, 171.2, 172.8 ppm. EI-MS: *m*/*z* (%) = 320 (4) [M$^{+}$·], 162 (78), 114 (31), 43 (100). $C_{17}H_{24}N_2O_4$ (320.38): calcd. C 63.73, H 7.55, N 8.74, O 19.98; found C 63.84, H 7.52, N 8.70.

**Removal of the Methyl Ester Group of Dipeptides 4a−c:**[22] Removal of the methyl ester group of the dipeptides **4a−c** (1 mmol) was carried out with the reagent system $AlCl_3$ (9 mmol)/*N*,*N*-dimethylaniline (15 mmol). Chromatographic purification of the crude reaction products afforded the respective *N*-Fmoc-dipeptides **6a−c** in high yields. The compound characterization data for **6b** fitted those observed for an authentic sample of *N*-Fmoc-valyl-leucine.[22]

**Compound 6a:** White solid (0.25 g, 78% yield), m.p. 167−170 °C. IR (KBr): ṽ = 3390, 2962, 1707, 1670, 1646, 1554, 1246, 761, 740 cm$^{-1}$. $^1$H NMR ([D$_6$]DMSO): δ = 0.73−0.89 [m, 6 H, $CH_2CH(CH_3)_2$], 1.41−1.70 [m, 3 H, $CH_2CH(CH_3)_2$], 2.92−3.02 (m, 2 H, $CH_2Ph$), 3.98−4.38 (m, 4 H, Fmoc-$CH_2$, Fmoc-CH, $CHCH_2Ph$), 4.72 [m, 1 H, $CHCH_2CH(CH_3)_2$], 7.08−8.29 (m, 15 H, OCONH, CHCONH, Ar-*H*) ppm. FAB$^+$ MS: *m*/*z* (%) = 501 (17) [M + H]$^+$, 179 (100), 178 (62), 165 (40). $C_{30}H_{32}N_2O_5$ (500.59): calcd. C 71.98, H 6.44, N 5.60, O 15.98; found C 72.09, H 6.40, N 5.57.

**Compound 6c:** White solid (0.31 g, 75% yield), m.p. 183−186 °C. IR (KBr): ṽ = 3408, 2961, 1692, 1656, 1254, 759, 740 cm$^{-1}$. $^1$H NMR ([D$_6$]DMSO): δ = 0.78−0.94 [m, 6 H, $CH(CH_3)_2$], 2.05 [m, 1 H, $CH(CH_3)_2$], 2.70−3.00 (m, 2 H, $CH_2Ph$), 4.03−4.31 [m, 4 H, Fmoc-$CH_2$, Fmoc-CH, $CHCH(CH_3)_2$], 4.37 (m, 1 H, $CHCH_2Ph$), 7.13−7.90 (m, 15 H, OCONH, CHCONH, Ar-*H*) ppm. FAB$^+$ MS: *m*/*z* (%) = 487 (13) [M + H]$^+$, 179 (100), 178 (64), 165 (29). $C_{29}H_{30}N_2O_5$ (486.56): calcd. C 71.59, H 6.21, N 5.76, O 16.44; found C 71.71, H 6.17, N 5.73.


[1] For a comprehensive review, see: E. Haslam, *Tetrahedron* **1980**, *36*, 2409−2434.
[2] R. W. Roeske, *The Peptides*, Academic Press, New York, **1981**, vol. 3, p. 102.
[3] J. Jones, *The Chemical Synthesis of Peptides*, Clarendon Press, Oxford, **1991**, p. 103.
[4] M. Bodanzski, *Principles of Peptide Synthesis*, Springer-Verlag, Berlin, **1984**, 2nd ed., p. 221.
[5] S. Liu, C. Dockendorff, S. D. Taylor, *Org. Lett.* **2001**, *3*, 1571−1574.
[6] B. Ye, T. R. Burke, *Tetrahedron Lett.* **1995**, *36*, 4733−4736.
[7] R. Pascal, R. Sola, *Tetrahedron Lett.* **1998**, *39*, 5031−5034.
[8] M. G. Organ, J. Xu, B. N'Zemba, *Tetrahedron Lett.* **2002**, *43*, 8177−8180.
[9] R. W. Jackson, *Tetrahedron Lett.* **2001**, *42*, 5163−5165.
[10] M. J. O'Donnell, F. Delgado, *Tetrahedron* **2001**, *57*, 6641−6650.
[11] Y-Q. Wu, D. C. Limburg, D. E. Wilkinson, M. J. Vaal, G. S. Hamilton, *Tetrahedron Lett.* **2000**, *41*, 2847−2849.
[12] G. M. Makara, G. R. Marshall, *Tetrahedron Lett.* **1997**, *38*, 5069−5072.
[13] C. W. West, M. A. Estiarte, D. H. Rich, *Org. Lett.* **2001**, *3*, 1205−1208.
[14] D. Filippov, E. K. Yeheskiely, G. A. van der Marel, G. I. Tesser, J. H. van Boom, *Tetrahedron Lett.* **1998**, *39*, 3597−3600.
[15] K. J. Hale, J. Cai, V. M. Delisser, *Tetrahedron Lett.* **1996**, *37*, 9345−9348.
[16] W.-C. Chen, M. D. Vera, M. M. Joullie, *Tetrahedron Lett.* **1997**, *38*, 4025−4028.
[17] M. Nishizawa, H. Yamamoto, K. Seo, H. Imagawa, T. Sugihara, *Org. Lett.* **2002**, *4*, 1947−1949.
[18] Y. Luo, M. A. Blaskovich, G. A. Lajoie, *J. Org. Chem.* **1999**, *64*, 6106−6111.
[19] P. Gomez-Martinez, A. M. Kimbonguila, F. Guibe, *Tetrahedron* **1999**, *55*, 6945−6960.
[20] J. M. Sanderson, P. Singh, C. W. G. Fishwick, J. B. C. Findlay, *J. Chem. Soc., Perkin Trans. 1* **2000**, 3227−3231.
[21] J. M. Caba, I. M. Rodriguez, I. Manzanares, E. Giralt, F. Albericio, *J. Org. Chem.* **2001**, *66*, 7568−7574.
[22] M. L. Di Gioia, A. Leggio, A. Le Pera, C. Siciliano, G. Sindona, A. Liguori, *J. Pept. Res.* **2004**, *63*, 383−387.
[23] A. Leggio, A. Liguori, A. Napoli, C. Siciliano, G. Sindona, *Eur. J. Org. Chem.* **2000**, 573−575.
[24] M. L. Di Gioia, A. Leggio, A. Liguori, A. Napoli, C. Siciliano, G. Sindona, *J. Org. Chem.* **2001**, *66*, 7002−7007.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