# Synthesis of *N*-{[(9*H*-Fluoren-9-yl)methoxy]carbonyl}-Protected (Fmoc) β-Amino Acids (= Homo-α-Amino Acids) by Direct Homologation

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The successful application of the *Arndt-Eistert* protocol starting from commercially available *N*-{[(9*H*-fluoren-9-yl)methoxy]carbonyl}-protected (Fmoc) α-amino acids leading to enantiomerically pure *N*-Fmoc-protected β-amino acids in only two steps and with high yield is reported.

**Introduction.** – Interest in the synthesis of enantiomerically pure β-amino acids (= homo-α-amino acids) has enormously increased in the last decade due to the pharmacological and biological effects shown by the latter in free form and incorporated in peptides [1]. Only recently, the synthesis and extraordinary properties of a hexapeptide consisting of β-amino acids have been published [2].

With respect to the synthesis of peptides *N*-{[(9*H*-fluoren-9-yl)methoxy]carbonyl}-protected (Fmoc) β-amino acids should be of great value since these derivatives could be used in the well-established solid-phase method, eventually also in computer-based peptide synthesis[1]). Furthermore, these compounds would also be of great interest in the combinatorial-chemistry strategy [4].

The homologation of commercially available *N*-Fmoc-protected α-amino acids following the *Arndt-Eistert* pathway is a very attractive route, especially since the rearrangement using *N*-[(*tert*-butoxy)carbonyl](Boc)- and *N*-[(benzyloxy)carbonyl](Z)-protected intermediates has been shown to proceed stereospecifically to a high degree in most cases [5]. Unfortunately, the standard conditions used therein do not seem to be ideally suited for preserving the *N*-Fmoc protection, as the crucial step involves the presence of an excess of strongly basic amines. An alternate, less straightforward route of homologation involves reduction, substitution, and hydrolysis and leads to *N*-Fmoc derivatives *via* *N*-Boc deprotection and subsequent *N*-Fmoc protection of the target compond [4].

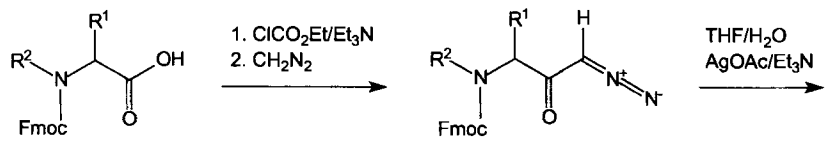
**Results and Discussion.** – We here report the successful application of the *Arndt-Eistert* protocol starting from the commercially available *N*-Fmoc-protected α-amino acids L-alanine (**1**), D-alanine (**2**), L-isoleucine (**3**), and L-proline (**4**) leading to enantiomerically pure *N*-Fmoc-protected β-amino acids by homologation in only two steps and with high yield[2]). The amino acids primarily investigated were selected for the following

[1]) While this paper was finalized, *Guichard* and *Seebach* published a note confirming the great value of these compounds in the construction of peptides containing nonproteinogenic amino acids [3].

[2]) Presented at the poster session of the '13. Wissenschaftl. Tagung der Österr. Pharmazeut. Gesellschaft,' Wien, April, 1997 [6].

reasons: *N*-Fmoc-L- and *N*-Fmoc-D-alanine (**1** and **2**, resp.) should provide us with samples of both enantiomers to enable the planned development of an HPLC-based method for the determination of the enantiomeric purity of β-amino acids [7]. *N*-Fmoc-L-Isoleucine (**3**) – due to the second stereogenic centre that is not involved in the applied reactions – has the essential prerequisites to investigate the outcome of the rearrangement by NMR spectroscopy. *N*-Fmoc-L-Proline (**4**), finally, should allow us to explore the limitations of the method and the applicability to heterocyclic amino acids.

*Scheme*

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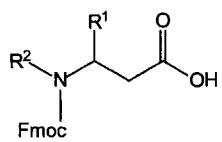
|---|---|---|---|---|---|
| **1** | $R^1$=Me, $R^2$=H; | Fmoc-Ala-OH | **5** | $R^1$=Me, $R^2$=H; | Fmoc-Ala-$CHN_2$ |
| **2** | $R^1$=Me, $R^2$=H; | Fmoc-D-Ala-OH | **6** | $R^1$=Me, $R^2$=H; | Fmoc-D-Ala-$CHN_2$ |
| **3** | $R^1$=(*S*)-*sec*-Bu, $R^2$=H; | Fmoc-Ile-OH | **7** | $R^1$=(*S*)-*sec*-Bu, $R^2$=H; | Fmoc-Ile-$CHN_2$ |
| **4** | $R^1$–$R^2$=$(CH_2)_3$; | Fmoc-Pro-OH | **8** | $R^1$–$R^2$=$(CH_2)_3$; | Fmoc-Pro-$CHN_2$ |

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|---|---|---|
| **9** | $R^1$=Me, $R^2$=H; | Fmoc-β-HAla-OH |
| **10** | $R^1$=Me, $R^2$=H; | Fmoc-β-HAla-OH |
| **11** | $R^1$=(*S*)-*sec*-Bu, $R^2$=H; | Fmoc-β-HIle-OH |
| **12** | $R^1$–$R^2$=$(CH_2)_3$; | Fmoc-β-HPro-OH |

The synthesis of the diazo ketone intermediates was easily performed by applying the mixed-anhydride method: the reaction with ethyl chloroformate (= ethyl carbonochloridate) was carried out at temperatures below $-20°$ avoiding an excess of $Et_3N$. After the addition of a freshly prepared solution of $CH_2N_2$ in $Et_2O$ at $< -20°$, the reaction mixture was allowed to warm up to 0° before workup. Thus, in all transformations investigated, the diazo ketones resulted in high yield and in enantiomerically pure form (see *Scheme* and *Exper. Part*). The structures of the yellow *N*-Fmoc-amino diazo ketones **5**–**8** were established by the characteristic strong absorption at 2100 $cm^{-1}$ in the IR spectra and by the outstanding signals in their NMR spectra: the most notable features are the $^1$H-resonances near 5 ppm (in $CDCl_3$) or 6 ppm (in $(D_6)$DMSO) (H–$CN_2$), the $^{13}$C-signal near 52 ppm ($CHN_2$), and the large $^1J$(H,C) coupling constant of *ca.* 200 Hz for $CHN_2$.

Rearrangement to the β-amino-acid derivatives can be initiated either by UV light ($\lambda \geq 254$ nm) or by silver salts, preferably complexed in $Et_3N$ [8]. Orienting small-scale

experiments led us to the decision to apply the Ag-induced reaction[3]). To minimize the *N*-Fmoc deprotection under the strongly basic conditions, the reaction mixture was actively warmed up from $-20°$ to *ca.* 0° and acidified by addition of 10% aqueous citric acid immediately after the rearrangement was complete. The desired *N*-Fmoc-protected β-amino acids **9**–**12** could be isolated in high yield after the usual workup procedure (see *Scheme* and *Exper. Part*). The structures of the homologated amino acids were established mainly by NMR spectroscopy, the most striking signals being due to the diastereotopic protons of the new $CH_2(\alpha)$ group (*AB* part of an *ABX* spin system).

The supposed retention of configuration in the β-amino acids was strongly suggested by the NMR spectra of *N*-Fmoc-L-β-homoisoleucine (**11**) in which any epimerization should be detectable *a priori* due to the presence of two stereogenic centres. Under standard conditions in $CDCl_3$, **11** shows various broadened signals of very low intensity, but on warming up to 60°, the spins do not exchange fast enough to simplify the spectrum; thus, the presence of the epimeric compound cannot be excluded. However, the $^1$H-NMR spectra of **11** recorded in ($D_6$)DMSO or ($D_6$)acetone exhibit a single set of data which suggest its enantiomeric purity, at least after recrystallization. Thus, the additional signals found for **11** in $CDCl_3$ solution may be due to the presence of conformational isomers. To further support this finding, we also converted the Fmoc-Ile-OH derived diazo ketone **7** into the methyl ester of **11** using a similar procedure. Both $^1$H- and $^{13}$C-NMR spectra gave no hints for the formation of stereoisomers during the rearrangement.

The stability of the *N*-Fmoc-protected β-amino acids **9**–**12** at elevated temperatures in ($D_6$)DMSO solution is limited, and, therefore, $^{13}$C-NMR data obtained at this temperature always showed signals of decomposition products (9-methylidene-9*H*-fluorene, deprotected amino acid) beside the expected signals. This problem required a special recording procedure in the case of the most unstable β-amino-acid derivative *N*-Fmoc-β-homoproline (**12**), which exhibited two sets of $^{13}$C-NMR data in $CDCl_3$ and in ($D_6$)DMSO solution at 30° that could not be brought to coalescence at higher temperatures. However, application of the new time-saving pulsed-field-gradient-supported 2D sequences enabled us to prove the exchange of the corresponding C-atoms of **12** and to completely assign the signals also at elevated temperatures (see *Fig.*).

Finally, comparison of the determined specific rotation of the diazo ketones derived from *N*-Fmoc-L-alanine (**1**) and *N*-Fmoc-D-alanine (**2**) and of the corresponding homologated amino acids confirm that the *Wolff* rearrangement proceeds stereospecifically.

**Conclusions and Perspectives.** – The *Arndt-Eistert* pathway is a fast and convenient way to synthesize *N*-Fmoc-protected β-amino acids[4]), at least at a laboratory scale[5]). The great and increasing number of commerically available *N*-Fmoc-protected α-amino

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[3]) Small-scale experiments monitored by TLC and NMR spectroscopy showed that the light-induced reaction led to mixtures containing olefinic by-products, presumably dimers of carbene intermediates. Nevertheless, the application of the 'Photo-*Wolff* conditions' should also lead to the desired enantiomerically pure products.

[4]) The methodology was also successfully applied to the orthogonally diprotected $N^{\alpha}$-Boc-$N^{\varepsilon}$-Fmoc-lysine [10].

[5]) For safety reasons, in our laboratory, all reactions involving diazomethane are performed at a scale not greater than 0.2M [11].

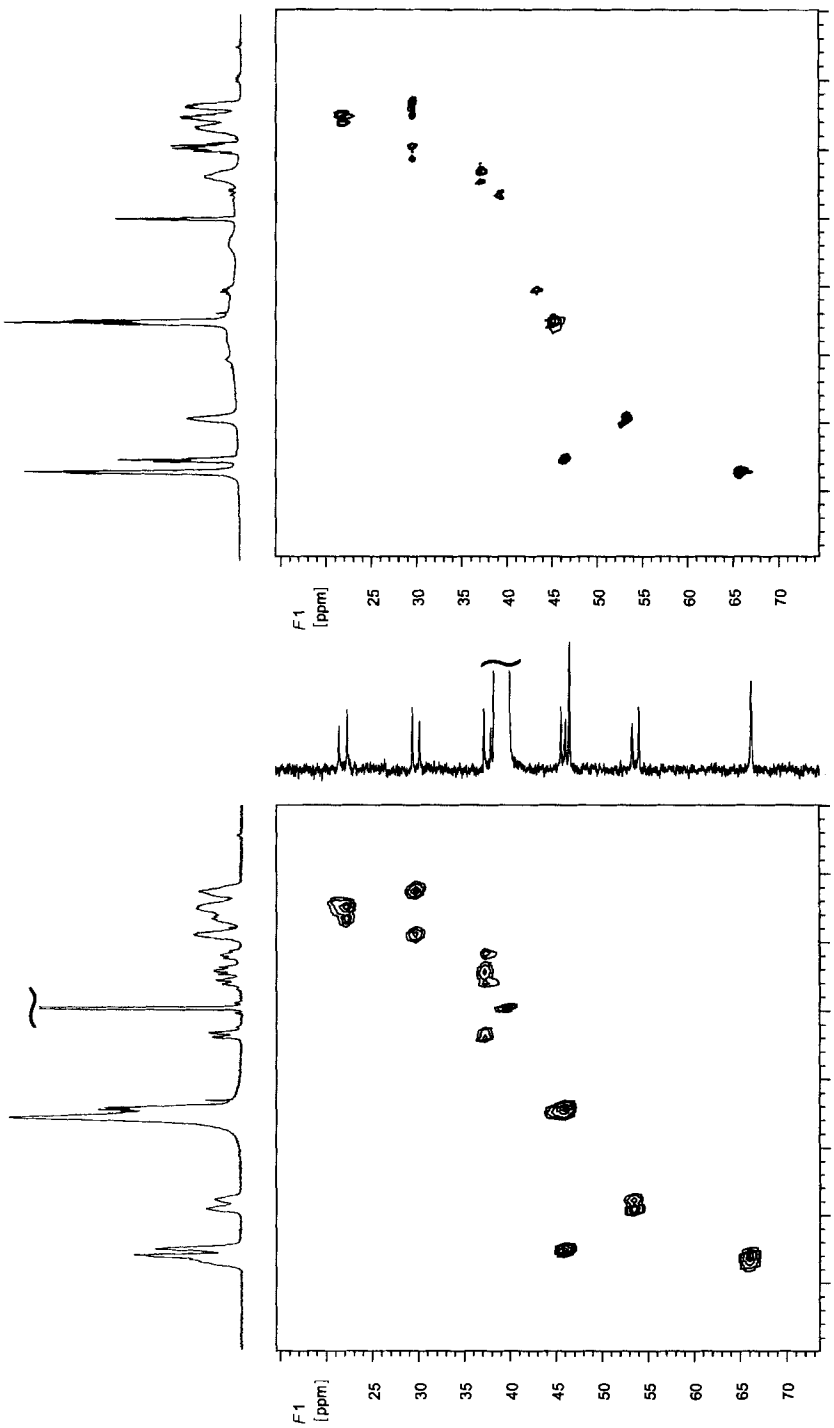


Figure. *Part of 2D plots of pulsed field gradient enhanced single-quantum coherence experiments of* **12** *at 30°* (left) *and 60°* (right). Data matrix size $2 \times 256 \times 1024$, 1 scan per $t_1$ value. Decoupling during acquisition was achieved with the use of the WURST decoupling sequence [9]. Shifted square sine-bell were used both in $t_1$ and $t_2$. 1D $^1$H-NMR at 500 MHz and $^{13}$C-NMR at 75 MHz.

acids makes this route a solid basis for the synthesis of β-amino-acid-based peptides either by the conventional solid-phase synthesis or by the recently developed methodology of *Seebach* and coworkers [12].

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## Experimental Part

*General.* Solvents were either used as purchased from *Fluka* (*puriss. p.a.* or *puriss.* quality) or distilled over appropriate drying agents. Amino-acid derivatives were purchased from *Senn.* TLC: *Macherey-Nagel Polygram® SIL G/UV*$_{254}$ anal. plates; detection either with UV or by spraying $Ce(SO_4)_2$ soln. and heating on a hot plate. M.p.: *Reichert* microscope with micro hot stage; uncorrected. Optical rotations: 10-cm, 1-ml cell at 20°; *Perkin-Elmer 141* polarimeter. IR Spectra: in $cm^{-1}$; *Galaxy 3000 FT* spectrometer, $CHCl_3$ soln. or KBr. NMR Spectra: chemical shifts $\delta$ in ppm and coupling constants $J$ in Hz, using the appropriate solvent as internal standard; *Varian Gemini 200* ($^1$H 200 MHz; $^{13}$C 50 MHz) or *Bruker AM 300* ($^1$H 300 MHz; $^{13}$C 75 MHz) or *Varian Unityplusm 500* ($^1$H, 500 MHz; $^{13}$C, 125 MHz) spectrometer; assignments based on DEPT spectra or 2D correlated spectroscopy (COSY, HMQC or HSQC). MS: *Finnigan MAT 95*; Cs gun, 20 kV, $2 \cdot 10^{-6}$ A; 3-nitrobenzyl alcohol (NOBA) matrix. Elemental analyses were performed at the Microanalytical Laboratory of the Institute of Physical Chemistry at the University of Vienna.

*N-Fmoc-Protected Amino Diazo Ketones: General Procedure (GP1).* A soln. of the *N*-Fmoc-protected α-amino acid in dry THF is cooled to −25° under Ar, $Et_3N$ (1.1 mol-equiv.) and ethyl chloroformate (1.1 mol-equiv.) are added subsequently with stirring while the temp. is kept below −20°. After 15 min, a freshly prepared $CH_2N_2$ soln. in dry $Et_2O$ [13][14] is added until the yellow colour of the soln. persists. The mixture is allowed to warm up to 0°, and stirring is continued for 2 to 12 h. Acidification with dil. acetic or citric acid and addition of an equal volume of $Et_2O$ is followed by washing the org. layer with $H_2O$, sat. $NaHCO_3$ soln., and brine. Drying ($Na_2SO_4$) and evaporation yields the crude diazo ketone which is further purified by suspending and stirring for 30 min in pentane and by recrystallization from the appropriate solvent.

*(3S)-1-Diazo-3-{{[(9H-fluoren-9-yl)methoxy]carbonyl}amino}butan-2-one* (**5**). From Fmoc-Ala-OH (**1**; 9.2 g, 29.5 mmol) according to *GP1*: 9.9 g (100%) of essentially pure **5** (by $^1$H-NMR). Recrystallization from *t*-BuOMe improved the purity of the material only slightly. Yellow crystals. M.p. 116°. For anal. purposes, the substance was recrystallized twice from *t*-BuOMe. $[\alpha]_D^{20} = -50.2$ ($c = 1.5$, DMF). IR ($CHCl_3$): 3434*w*, 3116*w*, 3011*m*, 2976*m*, 2894*w*, 2113*s*, 1719*s*, 1644*m*, 1504*s*, 1451*m*, 1386*m*, 1362*s*, 1331*w*, 1232*m*, 1147*w*, 1047*m*, 1032*m*, 877*w*. $^1$H-NMR ((D$_6$)DMSO, 300 MHz, 30°): 7.88 (*d*, 2 arom. H); 7.70 (br. *d*, 2 arom. H, NH (overlapping)); 7.41 (*t*, 2 arom. H); 7.32 (*t*, 2 arom. H); 5.95 (br. *s*, H–C(1)); 4.33 (*m*, $CH_2O$); 4.21 (*m*, H–C(9′)); 4.03 (*m*, H–C(3)); 1.19 (*d*, $J = 7.4$, Me). $^{13}$C-NMR ($CDCl_3$, 75 MHz, 30°): 195.54 (*s*, CO); 155.66 (*s*, NHCOO); 143.74, 140.70 (2*s*), 127.55, 126.97, 125.17, 125.10, 120.02 (5*d*, 12 arom. C); 65.45 (*t*, $CH_2O$); 53.83 (*d*, C(3)); 52.19 (*d*, C(1)); 46.69 (*d*, C(9′)); 16.84 (*q*, C(4)). FAB-MS: 337.1 (23.9, $[M + 2]^+$), 336.1 (100, $[M + 1]^+$), 335.1 (41.2, $M^+$). Anal. calc. for $C_{19}H_{17}N_3O_3 \cdot 0.15\ C_5H_{12}O$ ('348.59'): C 68.05, H 5.44, N 12.05; found: C 68.53, H 5.28, N 11.91.

*(3R)-1-Diazo-3-{{[(9H-fluoren-9-yl)methoxy]carbonyl}amino}butan-2-one* (**6**). From Fmoc-D-Ala-OH · $H_2O$ (**2**; 10.4 g, 31.6 mmol) according to *GP1*: 10.6 g (100%) of essentially pure **6** (by $^1$H-NMR). Yellow crystals. M.p. 118–119°. For anal. purposes, the substance was recrystallized from *t*-BuOMe. $[\alpha]_D^{20} = +50.1$ ($c = 0.9$, DMF). IR ($CHCl_3$): 3435*w*, 3116*w*, 3014*m*, 2977*m*, 2894*w*, 2113*s*, 1718*s*, 1645*m*, 1504*s*, 1451*m*, 1386*m*, 1362*s*, 1331*w*, 1232*m*, 1216*m*, 1147*w*, 1047*m*, 1031*m*, 876*w*. $^1$H-NMR ($CDCl_3$, 300 MHz, 26°): 7.75 (*d*, 2 arom. H); 7.57 (br. *m*, 2 arom. H); 7.38 (*t*, 2 arom. H); 7.30 (*t*, 2 arom. H); 5.39 (br. *d*, NH); 5.29 (br. *s*, H–C(1)); 4.42 (*m*, $CH_2O$); 4.42 (br. *m*, H–C(3)); 4.19 (*m*, H–C(9′)); 1.33 (*d*, $J = 6.8$, Me). $^{13}$C-NMR ((D$_6$)DMSO, 75 MHz, 30°): 195.66 (*s*, CO); 155.71 (*s*, NHCOO); 143.77, 140.75 (2*s*), 127.61, 127.03, 125.24, 125.17, 120.11 (6*d*, 12 arom. C); 65.49 (*t*, $CH_2O$); 53.88 (*d*, C(3)); 52.28 (*d*, C(1)); 46.71 (*d*, C(9′)); 16.89 (*q*, C(4)). FAB-MS: 337.1 (22.3, $[M + 2]^+$), 336.1 (100, $[M + 1]^+$), 335.1 (35.8, $M^+$). Anal. calc. for $C_{19}H_{17}N_3O_3$ (335.36): C 68.05, H 5.11, N 12.53; found: C 68.25, H 5.09, N 12.27.

*(3S,4S)-1-Diazo-3-{{[(9H-fluoren-9-yl)methoxy]carbonyl}amino}-4-methylhexan-2-one* (**7**). From Fmoc-Ile-OH (**3**; 25.0 g, 70.7 mmol) according to *GP1*. Recrystallization from THF/pentane 1:2 (*v*/*v*) gave 22.0 g (82%) of **7**. Yellow crystals. M.p. 136–137°. For anal. purposes, the substance was recrystallized twice from THF/pentane. $[\alpha]_D^{20} = -46.6$ ($c = 1$, DMF). IR (KBr): 3302*s*, 3082*m*, 3016*w*, 2965*m*, 2097*s*, 1692*s*, 1634*s*, 1537*m*, 1452*w*,

1389*m*, 1370*m*, 1321*w*, 1260*m*, 1238*w*, 1225*m*, 1028*m*. $^1$H-NMR ($CDCl_3$, 300 MHz, 26°): 7.75 (*d*, 2 arom. H); 7.57 (br. *d*, 2 arom. H); 7.39 (*t*, 2 arom. H); 7.30 (*td*, 2 arom. H); 5.35 (br. *d*, $J$ = 8.4, NH); 5.29 (br. *s*, H–C(1)); 4.42 (*m*, $CH_2O$); 4.20 (*m*, H–C(9′)); 4.13 (*m*, H–C(3)); 1.84 (*m*, H–C(4)); 1.44, 1.11 (2*m*, 2 H–C(5)); 0.93 (*d*, $J$ = 7.1, Me–C(4)); 0.90 (*t*, $J$ = 7.5, Me(6)). $^{13}$C-NMR ($CDCl_3$, 75 MHz, 26°): 193.23 (*s*, CO); 156.17 (*s*, MHCOO); 143.77, 141.33 (2*s*), 127.70, 127.07, 125.07, 125.00, 119.97 (5*d*, 12 arom. C); 66.79 (*t*, $CH_2O$); 62.28 (*d*, C(3)); 54.85 (*d*, C(1)); 47.27 (*d*, C(9′)); 37.62 (*d*, C(4)); 24.58 (*t*, C(5)); 15.64 (*q*, C(6)); 11.51 (*q*, (*Me*–C(4)). FAB-MS: 379.1 (24.2, $[M + 2]^+$), 378.1 (100, $[M + 1]^+$), 377.1 (41.3, $M^+$). Anal. calc. for $C_{22}H_{23}N_3O_3 \cdot 0.2\,C_5H_{12}$ ('391.88'): C 70.50, H 6.53, N 10.72; found: C 70.40, H 6.40, N 10.67.

*2-Diazo-1-{(2′S)-1-{[(9H-Fluoren-9-yl)methoxy]carbonyl}pyrrolidin-2-yl}ethan-1-one* (**8**). From Fmoc-Pro-OH (**4**; 9.9 g, 2.9 mmol) according to *GP1*: 9.55 g (90%) of essentially pure **8** (by $^1$H-NMR). Recrystallization from MeOH yielded 8.88 g (84%) of yellow crystals. M.p. 136–138°. $[\alpha]_D^{20} = -69.7$ ($c$ = 0.9, DMF). IR ($CHCl_3$): 3011*m*, 2985*w*, 2956*w*, 2110*s*, 1697*s*, 1642*s*, 1478*m*, 1451*s*, 1416*s*, 1356*s*, 1325*m*, 1122*m*. $^1$H-NMR ($CDCl_3$, 500 MHz, 26°): 7.75 (br. *t*, 2 arom. H); 7.60 (*m*, 1 arom. H); 7.53 (*t*, 1 arom. H); 7.38 (br. *t*, 2 arom. H); 7.30 (*t*, 2 arom. H); 5.28 (br. *s*, H–C(2) (conformer A)); 4.94 (br. *s*, H–C(2) (conformer B)); 4.55 (br. *m*, $CH_2O$ (conformer A), 1 H of $CH_2O$ (conformer B)); 4.39 (*m*, 1 H of $CH_2O$ (conformer B)); 4.30 (br. *m*, H–C(2′) (conformer B)); 4.23 (*t*, $J$ = 6.7, H–C(″) (conformer A)); 4.16 (*t*, $J$ = 5.2, H–C(9″) (conformer B)); 3.96 (br. *m*, H–C(2′) (conformer A)); 3.47 (*m*, $CH_2$(5′)); 2.15–1.95 (*m*, $CH_2$(3′)); 1.95–1.75 (*m*, $CH_2$(4′)). $^{13}$C-NMR ((D$_6$)DMSO, 75 MHz, 30°): 194.66 (*s*, CO); 153.99, 153.73 (2*s*, NHCOO); 143.72, 143.69, 140.68 (3*s*), 127.57, 127.04, 126.91, 125.03, 124.94, 120.01 (6*d*, 12 arom. C); 66.63, 66.49 (2*t*, $CH_2O$); 63.50, 62.95 (2*d*, C(2′)); 52.30 (*d*, C(2)); 46.97, 46.37 (2*t*, C(5′)); 46.64 (*d*, C(9″)); 30.62, 29.46 (2*t*, C(3′)); 23.75, 23.73 (2*t*, C(4′)). FAB-MS: 363.1 (26.1, $[M + 2]^+$), 362.1 (100, $[M + 1]^+$), 361.1 (44.9, $M^+$). Anal. calc. for $C_{21}H_{19}N_3O_3$ (361.40): C 69.79, H 5.30, N 11.63; found: C 70.08, H 5.24, N 11.49.

*N-Fmoc-Protected β-Amino Acids: General Procedure for the* Wolff *Rearrangement* (GP2). A soln. of AgOAc (0.11 mol-equiv.) in $Et_3N$ (3 mol-equiv.) is added to a soln. of the diazo compound in THF containing 10% $H_2O$ (*v*/*v*) at −15°. The mixture is actively warmed up within 10 min to −5 to 0° where the $N_2$ evolution starts; the temp. is kept below 5° until the reaction is complete (10 to 30 min, TLC monitoring). The mixture is filtered over *Celite*, the filtrate acidified by addition of 10% aq. citric acid soln. and diluted with an equal volume of AcOEt, and the org. layer washed with brine, dried ($Na_2SO_4$), and evaporated to yield the crude Fmoc-β-amino acid which can be further purified by recrystallization from the given solvent.

*(3S)-3-{{[(9H-Fluoren-9-yl)methoxy]carbonyl}amino}butanoic Acid* (= *N-{[(9H-Fluoren-9-yl)methoxy]carbonyl}*-L-*β-homoalanine*; *Fmoc-β-HAla-OH*; **9**). From **5** (1.0 g, 3 mmol) according to *GP2*: 0.9 g (92%) of essentially pure **9** (by $^1$H-NMR). Colourless crystals. M.p. 116–118°. For anal. purposes, the substance was recrystallized from EtOH/$H_2O$. $[\alpha]_D^{20} = +5.3$ ($c$ = 0.9, DMF). IR (KBr): 3322*m*, 3065*w*, 2961*m*, 1693*s*, 1536*s*, 1477*m*, 1450*s*, 1412*m*, 1378*m*, 1339*m*, 1261*s*, 1105*s*, 1086*s*, 1057*s*, 1032*m*, 977*w*, 935*w*, 758*m*, 737*s*. $^1$H-NMR ((D$_6$)DMSO, 300 MHz, 30°): 12.13 (br. *s*, COOH); 7.88 (*d*, 2 arom. H); 7.67 (br. *d*, 2 arom. H); 7.40 (br. *t*, 2 arom. H); 7.31 (*tt*, 2 arom. H); 7.21 (*d*, $J$ = 8.2, NH); 4.28 (*m*, $CH_2O$); 4.20 (*m*, H–C(9′)); 3.84 (*m*, H–C(3)); 2.35 (*AB* of *ABX*, $J$ = 15.4, 7.2, 6.6, $CH_2$(2)); 1.07 (*d*, $J$ = 6.7, Me). $^{13}$C-NMR ((D$_6$)DMSO, 75 MHz, 30°): 172.26 (*s*, COOH); 155.15 (*s*, NHCOO); 143.88, 143.81, 140.66 (3*s*), 127.51, 126.97, 125.07, 120.01 (4*d*, 12 arom. C); 65.12 (*t*, $CH_2O$); 46.70 (*d*, C(3)); 43.70 (*d*, C(9′)); 40.78 (*t*, C(2)); 20.39 (*q*, C(4)). FAB-MS: 327.1 (19.2, $[M + 2]^+$), 326.2 (100, $[M + 1]^+$), 325.1 (16.0, $M^+$). Anal. calc. for $C_{19}H_{19}NO_4$ (325.37): C 70.14, H 5.89, N 4.30; found: C 69.90, H 6.15, N 4.35.

*(3R)-3-{{[(9H-Fluoren-9-yl)methoxy]carbonyl}amino}butanoic Acid* (= *N-{[(9H-Fluoren-9-yl)methoxy]carbonyl}*-D-*β-homoalanine*; *Fmoc-D-β-HAla-OH*; **10**). From **6** (1.0 g, 2.67 mmol) according to *GP2*: 0.97 g (100%) of essentially pure **10** (by $^1$H-NMR). Colourless crystals. M.p. 115–118°. For anal. purposes, the substance was recrystallized from *t*-BuOMe. $[\alpha]_D^{20} = -4.7$ ($d$ = 1.1, DMF). IR (KBr): 3328*m*, 3066*w*, 3048*w*, 3040*w*, 3018*w*, 2965*w*, 2955*w*, 2949*w*, 2926*w*, 2853*w*, 1689*s*, 1540*m*, 1535*m*, 1451*w*, 1286*w*, 1262*m*, 1059*w*, 1028*w*, 737*m*. $^1$H-NMR ((D$_6$)DMSO, 300 MHz, 26°): 12.17 (br. *s*, COOH); 7.87 (*d*, 2 arom. H); 7.68 (br. *d*, 2 arom. H); 7.40 (br. *t*, 2 arom. H); 7.32 (*tt*, 2 arom. H); 7.26 (*d*, $J$ = 8.5, NH); 4.28 (*m*, $CH_2O$); 4.20 (*m*, H–C(9′)); 3.85 (*m*, H–C(3)); 2.35 (*AB* of *ABX*, $J$ = 15.4, 7.2, 6.6, $CH_2$(2)); 1.08 (*d*, $J$ = 7.0, Me). $^{13}$C-NMR ((D$_6$)DMSO, 75 MHz, 26°): 172.39 (*s*, COOH); 155.22 (*s*, NHCOO); 143.93, 143.87, 140.73 (3*s*), 127.58, 127.04, 125.13, 120.09 (4*d*, 12 arom. C); 65.17 (*t*, $CH_2O$); 46.75 (*d*, C(3)); 43.77 (*d*, C(9′)); 40.83 (*t*, C(2)); 20.47 (*q*, C(4)). FAB-MS: 327.1 (30.2, $[M + 2]^+$), 326.2 (100, $[M + 1]^+$), 325.1 (16.0, $M^+$). Anal. calc. for $C_{19}H_{19}NO_4 \cdot 0.96\,H_2O$ ('342.66'): C 66.60, H 6.15, N 4.09; found: C 66.55, H 5.58, N 3.98.

*(3R,4S)-3-{{[(9H-Fluoren-9-yl)methoxy]carbonyl}amino}-4-methylhexanoic Acid* (= *N-{[(9H-Fluoren-9-yl)methoxy]carbonyl}*-L-*β-homoisoleucine*; *Fmoc-β-HIle–OH*; **11**). From **7** (2.1 g, 6 mmol) according to *GP2*: 2.0 g (98%) of essentially pure **11** (by $^1$H-NMR). Colourless crystals. M.p. 98–100°. For anal. purposes,

the substance was recrystallized from EtOH/$H_2O$. $[\alpha]_D^{20} = +16.6$ ($c = 0.8$, DMF). IR (KBr): 3324*m*, 3067*w*, 2965*m*, 2932*w*, 2876*w*, 1698*s*, 1543*m*, 1451*m*, 1304*w*, 1262*m*, 1236*m* (sh), 1125*w*, 1042*w*, 739*m*. $^1$H-NMR ($(D_6)$DMSO, 500 MHz, 26°): 12.06 (br. *s*, COOH); 7.87 (*d*, 2 arom. H); 7.69 (*t*, 1 arom. H); 7.67 (*t*, 1 arom. H); 7.32 (*t*, 2 arom. H); 7.30 (*t*, 2 arom. H); 7.24 (*d*, $J = 8.7$, NH); 4.25 (*m*, $CH_2O$); 4.20 (*m*, H–C(9′)); 3.78 (*m*, H–C(3)); 2.31 (*AB* of *ABX*, $J = 15.4$, 9.4, 4.4, $CH_2$(2)); 1.47 (*m*, H–C(4)); 1.34 (*m*, 1 H–C(5)); 1.05 (*m*, 1 H–C(5)); 0.83 (*t*, $J = 7.3$, Me(6)); 0.79 (*d*, $J = 6.9$, Me–C(4)). $^{13}$C-NMR ($(D_6)$DMSO, 75 MHz, 30°): 172.88 (*s*, COOH); 155.47 (*s*, NHCOO); 143.95, 143.73, 140.65 (3*s*), 127.50, 126.92, 125.14, 119.99 (4*d*, 12 arom. C); 65.10 (*t*, $CH_2O$); 51.86 (*d*, C(3)); 46.76 (*d*, C(9′)); 38.32 (*d*, C(4)); 35.76 (*t*, C(2)); 24.91 (*t*, C(5)); 14.74 (*q*, *Me*–C(4)); 11.42 (*q*, C(6)). FAB-MS: 369.2 (25.6, $[M + 2]^+$), 368.2 (100, $[M + 1]^+$), 367.2 (7.6, $M^+$). Anal. calc. for $C_{22}H_{25}NO_4$ (367.44): C 71.91, H 6.86, N 3.81; found: C 71.97, H 7.09, N 3.90.

*Methyl (3*R*,4*S*)-3-{{[(9*H*-Fluoren-9-yl)methoxy]carbonyl}amino}-4-methylhexanoate* (= N-*{[(9*H*-Fluoren-9-yl)methoxy]carbonyl}*-L-*β-homoisoleucine Methyl Ester*; *Fmoc-β-HIle-OMe*). From **7** (3.0 g, 8 mmol) according to *GP2* with THF containing 40% of MeOH as solvent: 2.9 g (95%) of essentially pure Fmoc-β-HIle-OMe (by $^1$H-NMR). Colourless crystals. M.p. 120°. For anal. purposes, the substance was recrystallized from THF/$Et_2O$. $[\alpha]_D^{20} = 19.1$ ($c = 0.9$, DMF). IR (KBr): 3318*s*, 3085*w*, 3019*w*, 2962*s*, 2932*m*, 2888*w*, 2875*m*, 2857*w*, 1745*s*, 1693*s*, 1546*s*, 1464*m*, 1450*s*, 1417*w*, 1378*w*, 1345*m*, 1321*s*, 1309*s*, 1236*s*, 1218*m*, 1191*m*, 1176*m*, 1121*s*, 1080*w*, 1045*s*, 760*m*, 738*s*. $^1$H-NMR ($CDCl_3$, 300 MHz): 7.74 (*d*, 2 arom. H); 7.58 (*d*, 2 arom. H); 7.38 (*t*, 2 arom. H); 7.29 (*td*, 2 arom. H); 5.16 (*d*, $J = 9.0$, NH); 4.38 (*d*, $J = 7$, $CH_2O$); 4.21 (*t*, $J = 7$, H–C(9′)); 3.86 (*m*, H–C(3)); 3.66 (*s*, MeO); 2.51 (*AB* of *ABX*, $J = 15.5$, 6.9, 4.6, $CH_2$(2)); 1.61 (*m*, H–C(4)); 1.49 (*m*, 1 H–C(5)); 1.10 (*m*, 1 H–C(5)); 0.89 (*t*, $J = 7.4$, Me(6)); 0.87 (*d*, $J = 6.5$, Me–C(4)). $^{13}$C-NMR ($CDCl_3$, 75 MHz): 172.36 (*s*, COOMe); 155.90 (*s*, NHCOO); 144.01, 143.92, 141.31 (3*s*), 127.62, 126.99, 125.05, 119.92 (4*d*, 12 arom C); 66.55 (*t*, $CH_2O$); 52.47 (*d*, C(3)); 51.75 (*q*, MeO); 47.30 (*d*, C(9′)); 38.08 (*d*, C(4)); 36.25 (*t*, C(2)); 25.48 (*t*, C(5)); 15.27 (*q*, *Me*–C(4)); 11.36 (*q*, C(6)). FAB-MS: 383.1 (25.5, $[M + 2]^+$), 382.1 (100, $[M + 1]^+$), 381.1 (3.6, $M^+$). Anal. calc. for $C_{23}H_{27}NO_4$ (381.47): C 72.42, H 7.13, N 3.67; found: C 72.20, H 7.29, N 3.69.

*(2*S*)-1-{[(9*H*-Fluoren-9-yl)methoxy]carbonyl}pyrrolidine-2-acetic Acid* (*Fmoc-β-HPro-OH*, **12**). From **8** (1.0 g, 2.67 mmol) according to *GP2*: 0.9 g (93%) of essentially pure **12** (by $^1$H-NMR). Recrystallization from MeOH yielded 0.8 g (83%) of colourless crystals. M.p. 191–192°. $[\alpha]_D^{20} = -33.6$ ($c = 0.9$, DMF). IR ($CHCl_3$): 3068*m*, 3027*m*, 3008*m*, 2982*m*, 2955*m*, 1699*s*, 1451*s*, 1421*s*, 1355*m*, 1335*m*, 1121*m*. $^1$H-NMR ($(D_6)$DMSO, 500 MHz, 26°): 12.19 (br. *s*, COOH); 7.87 (br. *m*, 2 arom. H); 7.64 (*d*, 2 arom. H); 7.40 (br. *m*, 2 arom. H); 7.32 (br. *m*, 2 arom. H); 4.45–4.20 (br. *m*, $CH_2O$, H–C(9′)); 3.97 (*m*, H–C(2) (conformer A)); 3.89 (*m*, H–C(2) (conformer B)); 3.23 (*m*, $CH_2$(5)); 2.45 (*AB* of *ABX*, $CH_2$COOH (conformer A)); 2.20 (*AB* of *ABX*, $CH_2$COOH (conformer B)); 1.95 (br. *m*, 1 H of $CH_2$(3)); 1.83 (*m*, 1 H of $CH_2$(4)); 1.74 (*m*, 1 H of $CH_2$(4)); 1.63 (*m*, 1 H of $CH_2$(3)). $^{13}$C-NMR ($(D_6)$DMSO, 75 MHz, 26°): 172.47, 172.29 (2*s*, COOH); 153.67 (*s*, NHCOO); 143.88, 140.77 (2*s*), 127.60, 127.06, 125.05, 124.88, 120.09 (5*d*, 12 arom. C); 66.15 (*t*, $CH_2O$); 54.18, 53.48 (2*d*, C(2)); 46.74 (*d*, C(9′)); 46.30, 45.86 (2*t*, C(5)); 38.34, 37.58 (2*t*, $CH_2$COOH); 30.67, 29.90 (*t*, C(3)); 22.92, 22.02 (2*t*, C(4)). FAB-MS: 353.2 (23.4, $[M + 2]^+$), 352.2 (100, $[M + 1]^+$), 351.2 (5.9, $M^+$). Anal. calc. for $C_{21}H_{21}NO_4$ (351.40): C 71.78, H 6.02, N 3.99; found: C 72.04, H 6.29, N 4.04.

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