A Constrained Diketopiperazine as a New Scaffold for the Synthesis of Peptidomimetics

Jean-François Pons[a], Jean-Luc Fauchère[b], Frédéric Lamaty[a], Annie Molla[c], and René Lazaro*[a]

Laboratoire des Aminoacides, Peptides et Protéines CNRS ESA 5075, Université Montpellier II[a],
F-34095 Montpellier Cédex 05, France

Institut de Recherches Servier[b],
F-92150 Suresnes, France

Laboratoire d'Etude de l'Adhesion Cellulaire, CNRS UMR 5538[c],
Institut A. Bonniot, F-38000 Grenoble, France

Received October 9, 1997

Keywords: *N*-Alkylation / Diketopiperazine building block / Peptidomimetics / RGD analogues

As a new scaffold for peptidomimetic synthesis, a highly constrained bifunctional diketopiperazine, **4**, has been prepared by smooth *N*-alkylation with *tert*-butyl bromoacetate. As a first application, we describe herein the synthesis of new peptidomimetics of the Arg-Gly-Asp (RGD) sequence. The product **30**, which shows a selective platelet-aggregation inhibiting activity, can be used as a lead for the preparation of more potent products.

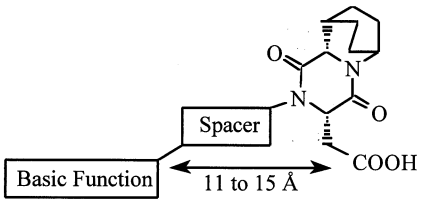
Introduction

2,5-Diketopiperazines (DKP), formed by cyclization of dipeptides, are interesting compounds because of their biological properties (stability to proteolysis)[1] and their rigid backbone, which can mimic a preferential peptide conformation. Moreover, since these molecules are easily synthesized, they have been used as building blocks in combinatorial chemistry[2][3]. The use of the bicyclic acid, 2-azabicyclo[2.2.2]octane-3-carboxylic acid (Abo) introduced an increased rigidity in the molecule c(Asp-Abo) **1**, which has been used to build efficient antagonists of tachykinin that exhibit antalgic activities comparable to those of morphine[4]. In order to permit the insertion of this building block in a sequence (and not only at the *N*-terminal position as in the case of tachykinin), we prepared a bifunctional derivative **4** containing an additional carboxylic acid group. We developed a non-racemizing, smooth alkylation of the available ring-nitrogen. This new building block was first used in the preparation of RGD sequence analogues that show higher specificity than the parent peptide in the inhibition of integrin-dependent cell aggregation. More specifically, we studied the inhibition of the interaction of $\alpha_{IIb}\beta_3$ integrin and fibrinogen on the one hand, and of that between $\alpha_5\beta_1$ integrin and fibrinogen or fibronectin on the other.

The general features of the synthesized RGD analogues[5] are described below. According to the literature, it is important to synthesize products having a carboxylic acid[6] and an ammonium[7][8][9] function within a distance of 11−15 Å[10]. Moreover, it has been shown that the basic function can be either a guanidine[7], which is the natural function of the RGD sequence, or an amine[8] such as piperidine or a benzamidine moiety[9].

We chose to synthesize derivatives in which the DKP scaffold and one of these groups are linked by a spacer of variable structure and length (Figure 1).

Figure 1. General scheme of derivatives studied



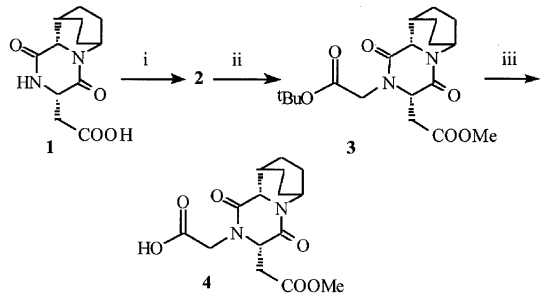
Results and Discussion

Synthesis of the N-Alkylated Diketopiperazine Scaffold: The DKP *cyclo*[(*S*)Asp-(*S*)Abo] (**1**) was used as the starting material[10]. After esterification of **1** with $SOCl_2$ in MeOH, the second function was introduced by *N*-alkylation of the DKP **2** (Scheme 1). The best results were obtained by addition of compound **2** to 1.1 equivalent of NaH in THF. 1.1 equivalent of *tert*-butyl bromoacetate was then added to afford the *N*-alkylated DKP **3** in 75% yield without racemization, along with 10% of recovered starting material. When more than 1.1 equivalent of NaH was used, racemization occurred and a diastereomeric mixture was detected by ^{1}H NMR and HPLC. Product **3** was crystallized and subjected to X-ray analysis, which confirmed the relative stereochemistry of the asymmetric centers of the DKP ring (Figure 3). The *tert*-butyl ester was then hydrolysed using trifluoroacetic acid (TFA) to give **4** in quantitative yield. This product was used as a building block for preparing the desired derivatives.

Synthesis of the Various Peptidomimetic Compounds: The first designed molecules incorporated a guanidine function

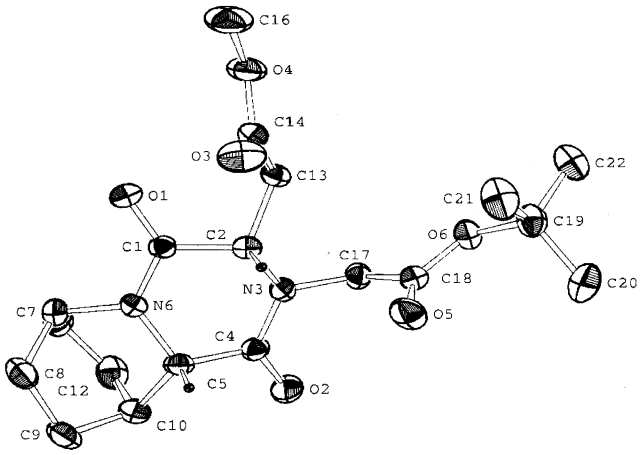
  1434−193X/98/0505−0853 $ 17.50+.50/0

Scheme 1. *N*-Alkylation of the DKP



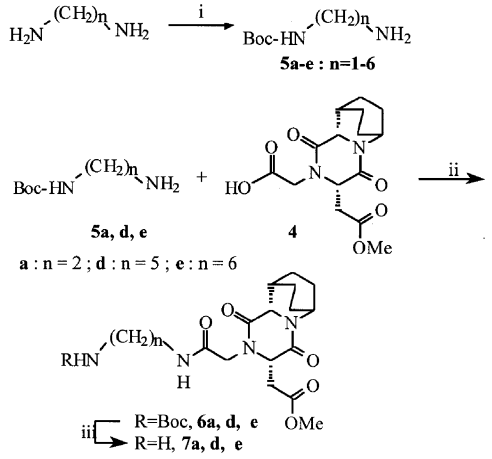
i) $SOCl_2$, MeOH, 25°C (quantitative yield); ii) NaH, $BrCH_2COOtBu$, THF, 25°C (75%); iii) TFA, 25°C (quantitative yield).

Figure 2. X-ray ORTEP of **3**



and a linear spacer such as $-NH-(CH_2)_n-NH$, $n = 2-6$, or $-NH-CH_2-CONH-(CH_2)_2-NH-$. The monoprotected diamines **5a**, **d**, **e** were coupled with compound **4** using DCC in the presence of DMAP and were deprotected with TFA in dichloromethane giving the products **7a**, **d**, **e** in yields of 90, 70 and 63%, respectively (Scheme 2).

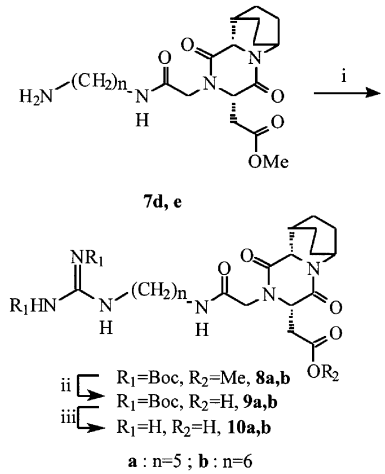
Scheme 2



i) Boc_2O, $CHCl_3$ (95%); ii) DCC, DMAP, dichloromethane (**6a** 90%, **6d** 70%, **6e** 63%,); iii) TFA, dichloromethane (quantitative yields).

Synthesis of **10a**, **b** *(Scheme 3):* The guanidine function was incorporated into the amines **7d** and **7e** as described by Kim and Qian[11]. Thus, by treatment with *N*,*N*′-bis(*tert*-butyloxycarbonyl)thiourea in the presence of mercuric chloride and triethylamine in DMF at 0°C, the products **8a** and **8b** were obtained in good yield. Deprotection of the methyl ester with NaOH in dioxane and hydrolysis of Boc with TFA in dichloromethane afforded the products **10a** and **10b** in 63 and 48% yield, respectively, based on **8a** and **8b**.

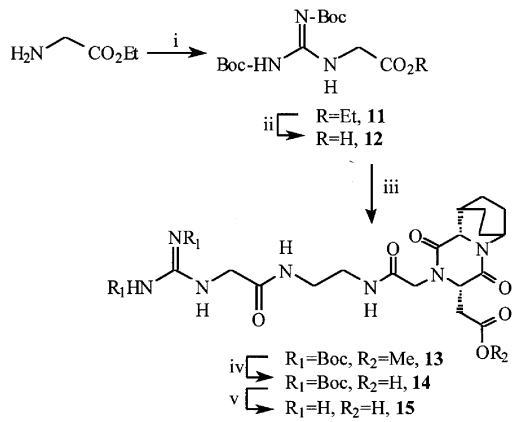
Scheme 3. Introduction of the guanidine function



i) Boc-NH−C(=S)−NH-Boc, $HgCl_2$, NEt_3, DMF (**8a** 76%, **8b** 88%); ii) NaOH (2 N), H_2O, dioxane (**9a** quantitative yield, **9b** 85%); iii) TFA, dichloromethane (**10a** 63%, **10b** 56%).

Synthesis of **15** *(Scheme 4):* The product **7a** was also functionalized with a guanidine group. In this case, an amide bond was introduced in the spacer. The glycine ethyl ester was guanidinated as previously described for **8a**, to give **11** in 72% yield, and was saponified with NaOH in 83% yield. The product **12** was coupled with **7a** using DCC and DMAP to afford **13** in 80% yield. Deprotection of the two functional groups afforded the product **15** in 58% yield.

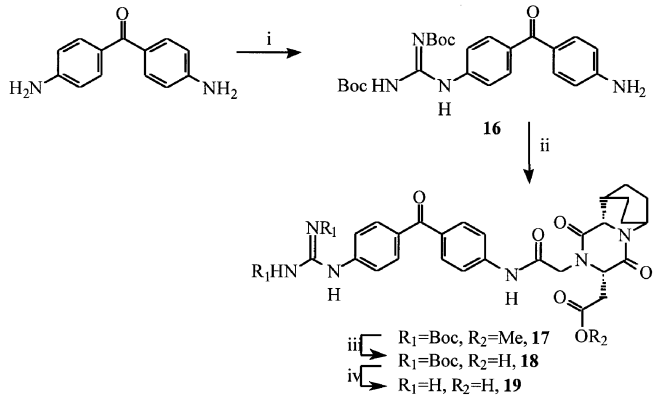
Scheme 4



i) Boc-NH−C(=S)−NH-Boc, $HgCl_2$, NEt_3, DMF (72%); ii) NaOH (2 N), H_2O, dioxane (83%); iii) **7a**, DCC, DMAP, dichloromethane (80%); iv) NaOH (2 N), H_2O, dioxane (75%); v) TFA, dichloromethane (78%).

Synthesis of **19** *(Scheme 5):* In order to introduce a more rigid spacer, 4,4′-diaminobenzophenone was converted to the monoguanidine derivative **16** in the presence of *N*,*N*′-bis(*tert*-butyloxycarbonyl)thiourea, mercuric chloride and pyridine in DMF at 0°C in 64% yield[12]. Coupling and deprotections afforded compound **19** in 33% yield based on **16**.

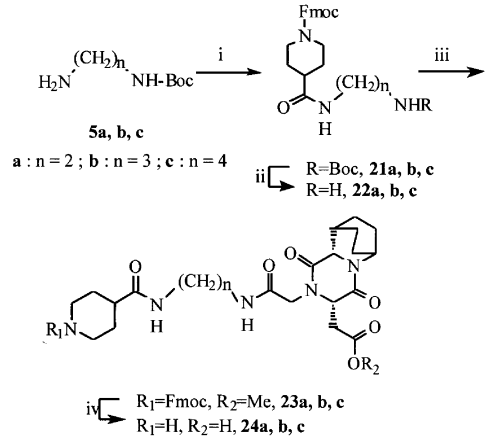
Scheme 5



i) Boc-NH−C(=S)−NH-Boc, $HgCl_2$, pyridine, DMF (64%); ii) **4**, DCC, DMAP, dichloromethane (44%); iii) NaOH (2 N), H_2O, dioxane (quantitative yield); iv) TFA, dichloromethane (76%).

Synthesis of **24a**, **b**, **c** *(Scheme 6):* In order to introduce a piperidine group instead of a guanidine, we started our synthesis from *N*-fluorenylmethyloxycarbonylisonipecotic acid (**20**), which was coupled with the monoprotected diamines **5a**, **b**, **c** using DCC in the presence of DMAP in dichloromethane to afford the products **21a**, **b**, **c** in 40, 47 and 50% yield, respectively. These compounds were then treated with TFA in dichloromethane, and coupled with **4** to afford the corresponding products **23a**, **b**, **c** in 53, 65 and 62% yield, respectively, based on **21a**, **b**, **c**. The deprotection of the two functional groups was achieved in a single step using NaOH in dioxane, furnishing the products **24a**, **b**, **c** in 53, 67 and 65% yield, respectively.

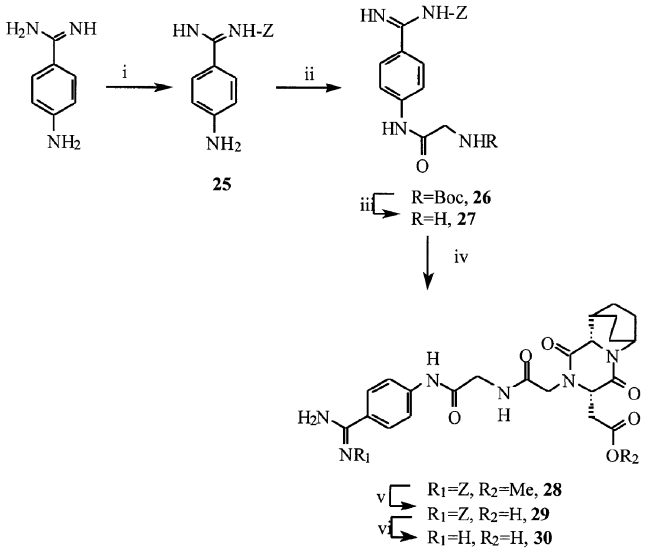
Scheme 6



i) *N*-Fmoc isonipecotic acid **20**, DCC, DMAP, dichloromethane (**21a**−**c** 40%, 47%, 50%, resp.); ii) TFA, dichloromethane (**22a**−**c** in quantitative yield, 80%, 92%, resp.); iii) **4**, BOP, DIEA, dichloromethane (**23a**−**c** 53%, 65%, 62%, resp.); iv) NaOH (2 N), H_2O, dioxane (**24a**−**c** 53%, 67%, 65%, resp.).

Synthesis of **30** *(Scheme 7):* The latter of the above compounds is a benzamidine derivative. The amidine function of the 4-aminobenzamidine was first protected by the action of NaOH and benzyl chloroformate in THF to afford **25** in 50% yield.[13] This product was reacted with the anhydride of *N*-*tert*-butyloxycarbonylglycine in the presence of DMAP in dichloromethane to give **26** in 42% yield.[14] The amine was deprotected with TFA in 74% yield and coupled with **4** to afford **28** in 31% yield. The saponification of the methyl ester was carried out as described above for **6**, giving a 73% yield, and subsequent hydrogenolysis afforded **30** in 49% yield.

Scheme 7



i) ZCl, NaOH, THF, H_2O (50%); ii) $(Boc\ Gly)_2O$, DMAP, dichloromethane (42%); iii) TFA, dichloromethane (74%); iv) DCC, DMAP, dichloromethane (31%); v) NaOH, dioxane, H_2O (73%); vi) $H_2/Pd(OH)_2/C$, EtOH (49%).

Biological Tests: Assays of inhibition of platelet aggregation mediated by fibrinogen were performed according to Zucker.[13] Platelet-rich plasma (PRP) was prepared by centrifugation (200g, 10 min) of citrated arterial dog blood. PRP was incubated for 10 min with a known concentration of the product to be tested. Thereafter, ADP (10 μM) was added and the aggregating effect was evaluated photometrically in a CHRONOLOG aggregometer (Coulotronics, France). The anti-aggregating activity was compared to that of the compound BIBU 52[14] (Figure 3 and Table 1), a highly active non-peptide fibrinogen-receptor antagonist.

Inhibition of the cell adhesion was measured on HEL (human erythroleukemia) cells (erythroblastic cell line developed from a patient who contracted leukemia after treatment for a solid tumor[15]). These cells were shown to grow in suspension, but to become adherent in the presence of the matrix proteins fibronectin (through $\alpha_5\beta_1$ integrin binding) or fibrinogen (through $\alpha_{IIb}\beta_3$ integrin binding)[16]. The

Figure 3. Structure of BIBU 52

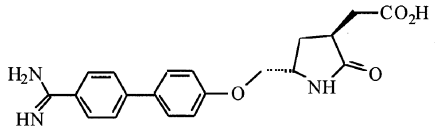


Table 1. Test of inhibition of platelet aggregation

Compound	IC_{50} (μM)[a]
10a, **10b**, **15**, **19**, **24a**, **24b**, **24c**	>100
30	18.0 ± 3.5
BIBU 52	0.110 ± 0.007

[a] Inhibition concentration in a dog PRP induced by ADP

cells were incubated with various concentrations of the products. The cell adhesion was measured after 45 min of incubation at 37°C in microtiter plates coated with matrix proteins. The wells were washed with PBS and adherent cells were counted after the Proméga reactant had been added. The optical density (OD) at 490 nm was measured following the manufacturer's instructions and calibrated with the OD of BSA alone. Experiments were performed at least in triplicate. A peptide containing the RGD sequence was taken as a positive standard, another containing RGE as a negative standard (Table 2). All these tests were performed in triplicate.

Table 2. Test of inhibition of cell adhesion

Compound	Fibrinogen IC_{50} (μM)	Fibronectin IC_{50} (μM)
10a, **10b**, **15**, **19**, **24a**, **24b**, **24c**	inactive	inactive
30	27 ± 7	>1000
GRGDS	1.8 ± 0.3	2 ± 0.5
GRGES	inactive	inactive

The results show that the compounds bearing a guanidine (**10a**, **10b**, **15**, **19**) or an amine (**24a**−**c**) function have no inhibitory activity on the fibrinogen or the fibronectin. On the contrary, the benzamidine derivative (**30**) exhibits an inhibitory effect on fibrinogen binding, but not on that of fibronectin. Thus, this compound has a specific effect on the interaction between the fibrinogen and its receptor GP IIb/IIIa located on the membrane surface of both the activated platelets and the HEL cells. This selectivity may be related to the presence of a benzamidine instead of a guanidine moiety[9] and/or to the higher rigidity of the constrained DKP compared to the linear RGDS peptide.

Conclusion

Smooth alkylation of the single free nitrogen atom of the diketopiperazine of Asp and Abo allows access to a series compounds in which the aspartyl residue is constrained in a heterocyclic ring. As a first application, these compounds were designed to act as mimetics of the tripeptide RGD. Each analogue was tested as an inhibitor of platelet aggregation and of cell adhesion. Compound **30** showed a modest but selective inhibition of fibrinogen binding. This new structure may be useful as a lead for the synthesis of new, more potent, selective RGD mimetics. Furthermore, we intend to use this new constrained DKP scaffold to prepare new mimetics of other biologically active peptides.

The authors wish to thank *Dr. T. J. Verbeuren (Institut de Recherches Servier, Suresnes)* for performing the dog platelet aggregation assays and *Dr. M. Pierrot (Université de Marseille III)* for the X-ray-spectroscopic analyses.

Experimental Section

General. − Solvents and Reagents: Unless otherwise stated, commercially available solvents and reagents were used. Dichloromethane, ethyl acetate and tetrahydrofuran were purified prior to use. − *NMR Spectroscopy:* NMR spectra were recorded on a Bruker AC-250 or WM 360 instrument in different solvents such as $CDCl_3$, CD_3OD or [D_6]DMSO. Chemical shifts are reported in δ values with TMS as an internal reference and splitting patterns are indicated as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet). − *Mass Spectrometry:* Molecular weights were determined by fast-atom bombardment mass-spectrometry (FAB MS) using a matrix of *m*-nitrobenzyl alcohol (NBA) or thioglycerol (GT) on Jeol JMS DX100 or DX300 spectrometers. High-resolution mass-spectrometry (HRMS) was employed for new compounds. − *Other Analytical Methods:* Melting points were determined using a Büchi capillary-tube melting-point apparatus and are not corrected. TLC was performed on silica gel (Merck™ 60F254) using appropriate solvent systems as eluents. Reaction products were visualized by UV fluorescence (254 nm) or by exposure to iodine vapour. The purity of products was checked by reversed-phase HPLC on a Waters Associates instrument equipped with two 510 model pumps. The absorbance was measured at 214 nm. The flow rate was 1 ml/min. in a Nucleosil 5-C18 column (10 μm, 25 × 250 mm) for semi-preparative HPLC.

General Procedure for Coupling an Amine and a Carboxylic Acid Derivative. − Method A: 1 mmol of the amine and 1 mmol of the carboxylic acid were dissolved in 10 ml of dichloromethane. At 0°C, 1.1 mmol of DCC and 0.1 mmol of DMAP were added. The reaction mixture was stirred for 15 h at room temperature. The solvent was then evaporated, the residue was redissolved in AcOEt, and the resulting solution was washed sequentially with saturated $NaHCO_3$ solution, H_2O, and 1 N HCl. The organic layer was dried over $MgSO_4$ and the solvent was evaporated. The residue was purified by column chromatography.

Method B: 1 mmol of the amine and 1 mmol of the carboxylic acid were dissolved in 10 ml of dichloromethane. At 0°C, 1.1 mmol of BOP and 3 mmol of DIEA were added. The reaction mixture was stirred for 15 h at room temperature. The solvents were then evaporated, the residue was redissolved in 20 ml of AcOEt, and the resulting solution was washed sequentially with saturated $NaHCO_3$ solution, H_2O, and 1 N HCl. The organic layer was dried over $MgSO_4$ and the solvent was evaporated.

General Procedure for the Removal of N-tert-Butyloxycarbonyl: 1 mmol of the protected compound was dissolved in 4 ml of dichloromethane. 1 ml of TFA was then added and the solution was stirred for 6 h at room temperature. The solvents were removed by evaporation.

General Procedure for the Removal of N-Benzyloxycarbonyl: 1 mmol of the benzyloxycarbonyl- (Z-)protected product was dis-

solved in 10 ml of EtOH. 200 mg of $Pd(OH)_2/C$ was added and the mixture was stirred for 6 h at room temperature under a hydrogen atmosphere (1 atm). The mixture was subsequently filtered through Celite and the filtrate was concentrated to dryness.

General Procedure for the Saponification of a Methyl or Ethyl Ester: 1 mmol of the methyl or ethyl ester was dissolved in 10 ml of a mixture of dioxane/water (2:1). 0.6 ml of NaOH (2 N) was added at 0°C. The reaction mixture was stirred for 6 h at room temperature, and then acidified to pH 3 with HCl (1 N) and extracted with AcOEt. The combined organic layers were dried over $MgSO_4$ and the solvent was evaporated.

General Procedure for Guanidination of an Amine[11]: 2.5 mmol of thiourea was dissolved in 50 ml of THF. At 0°C, 11.3 mmol of NaH was added and the mixture was stirred at this temperature for 5 min, then for 10 min at room temperature, and then again cooled to 0°C. 5.5 mmol of Boc_2O was added and the mixture was stirred for 30 min at 0°C and for 2 h at room temperature. After cooling to 0°C once more, the reaction was quenched by the addition of saturated $NaHCO_3$ solution, the organic phase was separated, and washed with brine. The aqueous phase was extracted with AcOEt. The combined organic layers were dried over $MgSO_4$ and the solvent was evaporated. The product was crystallized from diethyl ether to afford 550 mg of *N,N'*-bis(*tert*-butyloxycarbonyl)-thiourea (80%). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.60 (s). – MS (NBA): *m/z* = 277 ([M + H]$^+$). – M.p. 120–122°C (ref.[11] 124–127°C). – R_f = 0.92 (dichloromethane/AcOEt, 4:1).

The amine (1.2 mmol) was then treated with this reagent (1.2 mmol) in 2 ml of DMF. The solution was cooled to –10°C and 0.6 ml of NEt_3 and 1.2 mmol of $HgCl_2$ were added. The reaction mixture was stirred for 2 h at –10°C and for 1 h at room temperature. Then, 20 ml of AcOEt was added and the mixture was filtered through Celite. The filtrate was washed with brine then with 0.5 N HCl. The organic layer was dried over $MgSO_4$ and the solvent was evaporated.

(3S,6S)-1,4-Diaza-7,10-ethano-3-methylcarboxymethyl-2,5-dionebicyclo[4.4.0]decane (**2**): To a suspension of **1** (500 mg, 2 mmol) in 5 ml of MeOH, 0.29 ml (4 mmol) of thionyl chloride was slowly added at –10°C. The mixture was stirred for 3 h at room temperature. The solvents were then removed to give 530 mg of a white solid (quantitative yield). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.40–1.75 (m, 8 H), 2.49 (m, 1 H), 2.57 (dd, 1 H, *J* = 9 Hz, *J* = 17 Hz), 3.19 (dd, 1 H, *J* = 4 Hz, *J* = 17 Hz), 3.68 (s, 3 H), 3.98 (m, 1 H), 4.26 (m, 1 H), 4.40 (m, 1 H), 6.38 (m, 1 H). – MS (NBA): *m/z* = 267 ([M + H]$^+$). – M.p. 112–114°C. – $[\alpha]_D^{25}$ = –16 (*c* = 2, MeOH). – R_f = 0.37 (dichloromethane/AcOEt, 1:1).

(3S,6S)-1,4-Diaza-4-tert-butylcarboxymethyl-7,10-ethano-3-methylcarboxymethyl-2,5-dionebicyclo[4.4.0]decane (**3**): To a stirred suspension of NaH (53 mg, 2.2 mmol) in 15 ml of THF at –10°C under argon atmosphere, was added 530 mg of **2** (2 mmol). The solution was stirred for 10 min at –10°C, and then 0.36 ml of *tert*-butyl bromoacetate (2.2 mmol) was added. The reaction mixture was stirred for 8 h at room temperature and then acidified with 5 ml of 1 N HCl. The phases were separated and the aqueous layer was extracted with AcOEt. The combined organic layers were dried over $MgSO_4$ and the solvent was evaporated. The residue was purified by column chromatography (dichloromethane/AcOEt, 1:1) yielding 570 mg (75%) of a white powder, which could be crystallized from AcOEt. – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.43 (m, 9 H), 1.55–1.79 (m, 8 H), 2.59 (m, 1 H), 2.80 (dd, 1 H, *J* = 5 Hz, 16 Hz), 3.00 (dd, 1 H, *J* = 7 Hz, *J* = 16 Hz), 3.64 (d, 1 H, *J* = 18 Hz), 3.70 (s, 3 H), 4.07 (m, 1 H), 4.41 (m, 1 H), 4.55 (d, 1 H, *J* = 18 Hz), 4.59 (m, 1 H). – MS (NBA): *m/z* = 381 ([M + H]$^+$). – M.p. 146–148°C. – $[\alpha]_D^{25}$ = –26.1 (*c* = 1, MeOH). – R_f = 0.75 (dichloromethane/AcOEt, 1:1). – $C_{19}H_{28}O_6N_2$ (380.4): C 59.99, N 7.36, H 7.42; found C 60.09, N 7.55, H 7.57.

(3S,6S)-1,4-Diaza-4-carboxymethyl-7,10-ethano-3-methylcarboxymethyl-2,5-dionebicyclo[4.4.0]decane (**4**): 570 mg (1.5 mmol) of **3** was dissolved in 4 ml of TFA at 0°C. The reaction mixture was stirred for 1 h at 0°C and then for 3 h at room temperature. The solvents were removed under reduced pressure to afford 490 mg of a white powder (quantitative yield). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.47–1.85 (m, 8 H), 2.60 (m, 1 H), 2.80 (dd, 1 H, *J* = 5 Hz, *J* = 17 Hz), 3.12 (dd, 1 H, *J* = 7 Hz, *J* = 17 Hz), 3.57 (s, 3 H), 3.65 (d, 1 H, *J* = 18 Hz), 4.09 (m, 1 H), 4.41 (m, 1 H), 4.58 (t, 1 H), 4.69 (d, 1 H, *J* = 18 Hz), 9.45 (m, 1 H). – MS (GT): *m/z* = 325 ([M + H]$^+$). – m.p. 58–60°C. – R_f = 0.6 (dichloromethane/AcOEt, 1:1).

General Synthesis of the N-tert-Butyloxycarbonyldiamines **5**: 22 mmol of diamine was stirred in 100 ml of $CHCl_3$. 960 mg of $(Boc)_2O$ (4.4 mmol) dissolved in 50 ml of $CHCl_3$ was added dropwise at 0°C. The reaction mixture was stirred for 2 h at room temperature. The solution was filtered and the filtrate was concentrated. The oily residue was diluted with AcOEt and washed with brine. The aqueous layer was extracted with AcOEt. The organic layers were dried over $MgSO_4$ and the solvent was evaporated.

1,2-Diamino-N-tert-butyloxycarbonylethane (**5a**): 600 mg of **5a** was obtained as a colourless oil (85%). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.38 (s, 9 H), 1.60 (m, 2 H), 2.75 (t, 2 H, *J* = 6.5 Hz), 3.09 (q, 2 H, *J* = 6.5 Hz), 5.13 (m, 1 H). – MS (GT): *m/z* = 161 ([M + H]$^+$). – R_f = 0.45 (MeOH/NH_4OH, 99:1).

1,3-Diamino-N-tert-butyloxycarbonylpropane (**5b**): 660 mg of **5b** was obtained as a colourless oil (86%). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.36 (s, 9 H), 1.56 (q, 2 H, *J* = 6.5 Hz), 2.26 (m, 2 H), 2.71 (t, 2 H, *J* = 6.5 Hz), 3.14 (q, 2 H, *J* = 6 Hz), 5.09 (m, 1 H). – MS (GT): *m/z* = 175 ([M + H]$^+$). – R_f = 0.25 (MeOH/NH_4OH, 99:1).

1,4-Diamino-N-tert-butyloxycarbonylbutane (**5c**): 750 mg of **5c** was obtained as a colourless oil (90%). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.32 (m, 13 H), 2.78 (m, 2 H), 2.95 (m, 2 H), 3.76 (m, 1 H), 4.55 (m, 2 H). – MS (NBA): *m/z* = 189 ([M + H]$^+$). – R_f = 0.35 (MeOH/NH_4OH, 9:1).

1,5-Diamino-N-tert-butyloxycarbonylpentane (**5d**): 870 mg of **5d** was obtained as a colourless oil (98%). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.37 (m, 15 H), 2.62 (m, 2 H), 3.03 (m, 2 H), 3.67 (m, 1 H), 4.54 (m, 2 H). – MS (NBA): *m/z* = 203 ([M + H]$^+$). – R_f = 0.41 (MeOH/NH_4OH, 9:1).

1,6-Diamino-N-tert-butyloxycarbonylhexane (**5e**): 950 mg of **5e** was obtained as a slightly coloured oil (quantitative yield). – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.30–1.55 (m, 17 H), 2.58 (m, 2 H), 3.15 (m, 2 H), 3.42 (m, 1 H), 4.58 (m, 2 H). – MS (NBA): *m/z* = 217 ([M + H]$^+$). – R_f = 0.68 (MeOH/NH_4OH, 9:1).

8a: According to general coupling procedure A, **6d** was obtained in 70% yield from **5d** and **4**, and was then deprotected to afford **7d** in quantitative yield. **7d** was guanidinated following the general procedure to afford **8a** in 76% yield as a light-yellow oil. – ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.42–1.73 (m, 32 H), 2.55 (m, 1 H), 2.89–3.18 (m, 6 H), 3.64 (s, 3 H), 3.74–3.80 (d, 1 H, *J* = 16 Hz), 4.00–4.01 (m, 2 H), 4.19–4.26 (d, 1 H, *J* = 16 Hz), 4.39 (m, 1 H), 6.26 (m, 1 H), 8.24 (m, 1 H), 11.42 (m, 1 H). – MS (GT): *m/z* = 651 ([M + H]$^+$). – R_f = 0.24 (dichloromethane/AcOEt, 1:1).

8b: According to general coupling procedure A, **6e** was obtained in 63% yield from **5e** and **4**, and was then deprotected to afford

7e in quantitative yield. **7e** was guanidinated following the general procedure to afford **8b** in 88% yield as a light-yellow oil. − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.10−1.73 (m, 34 H), 2.55 (m, 1 H), 2.94−3.45 (m, 6 H), 3.66 (s, 3 H), 3.74−3.80 (d, 1 H, *J* = 16 Hz), 4.03−4.05 (m, 1 H), 4.19−4.26 (d, 1 H, *J* = 16 Hz), 4.38 (m, 1 H), 4.46 (m, 1 H), 6.25 (m, 1 H), 8.24 (m, 1 H), 11.42 (m, 1 H). − MS (NBA): *m/z* = 665 ([M + H]$^+$). − R_f = 0.43 (dichloromethane/AcOEt, 1:1).

10a: Deprotection of **8a** followed by purification by HPLC afforded **10a** as a light-yellow powder in 63% overall yield. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.27−1.83 (m, 14 H), 2.48 (m, 1 H), 2.79 (dd, 1 H, *J* = 4 Hz, *J* = 16 Hz), 2.97 (dd, 1 H, *J* = 7 Hz, *J* = 16 Hz), 3.12−3.18 (m, 4 H), 4.02 (dd, 1 H, *J* = 4 Hz, *J* = 7 Hz), 4.24−4.34 (m, 4 H). − ($C_{20}H_{33}N_6O_5$): HRMS (NBA): *m/z* = 437.2525 (for 437.2512). − M.p. 166−168°C. − HPLC: R_t = 6.3 min (H_2O/CH_3CN, 4:1). − $[\alpha]_D^{25}$ = +9.1 (*c* = 1, MeOH).

10b: Deprotection of **8b** followed by purification by HPLC afforded **10b** as a light-yellow powder in 48% overall yield. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.26−2.03 (m, 16 H), 2.51 (m, 1 H), 2.7 (m, 2 H), 3.19−3.33 (m, 5 H), 4.10−4.17 (m, 1 H), 4.33−4.41 (m, 3 H). − ($C_{21}H_{35}N_6O_5$): HRMS (NBA): *m/z* = 451.2702 (for 451.2669). − M.p. 96−98°C. − HPLC: *R*t = 20.2 min (H_2O/CH_3CN, 9:1). − $[\alpha]_D^{25}$ = +9.7 (*c* = 1, MeOH).

Ethyl 2-[N,N'-Bis(tert-butyloxycarbonyl)guanidino]ethanoate (**11**): 170 mg of glycine ethyl ester hydrochloride (1.2 mmol) was guanidinated according to the general procedure to afford 250 mg of **11** (72%) as white crystals. − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.12 (t, 3 H, *J* = 7 Hz), 1.43 (s, 9 H), 1.47 (s, 9 H), 4.15 (m, 2 H), 4.20 (m, 2 H), 8.83 (m, 1 H), 11.40 (m, 1 H). − MS (NBA): *m/z* = 346 ([M + H]$^+$). − M.p. 96−98°C. − R_f = 0.45 (dichloromethane/AcOEt, 1:1).

13: Compound **11** was saponified to give **12** (83%), which was coupled with **7a** according to the coupling procedure B to afford **13** in 80% yield as a white foam, which was not further purified. − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.44 (s, 9 H), 1.46 (s, 9 H), 1.52−1.76 (m, 8 H), 2.56 (m, 1 H), 3.00 (m, 2 H), 3.04−3.30 (m, 4 H), 3.66 (s, 3 H), 3.70 (m, 1 H), 3.99 (m, 1 H), 4.17 (m, 2 H), 4.37 (m, 2 H), 4.40 (m, 1 H), 4.53 (m, 1 H), 6.86 (m, 1 H), 8.85 (m, 1 H), 11.31 (m, 1 H). − MS (NBA): *m/z* = 666 ([M + H]$^+$). − M.p. 112−114°C. − R_f = 0.35 (dichloromethane/AcOEt, 1:1).

15: Compound **13** was saponified to give **14** in 75% yield, which was then *N*-deprotected. The product **15** was crystallized from AcOEt (58% based on **13**). − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.49−1.75 (m, 8 H), 2.40 (m, 1 H), 2.49−2.84 (m, 6 H), 3.50 (m, 2 H), 3.66 (d, 1 H), 3.70 (m, 2 H), 3.96 (m, 1 H), 4.21 (m, 1 H). − MS (NBA): *m/z* = 452 ([M + H]$^+$). − HRMS ($C_{19}H_{30}N_7O_6$): 452.2253 (for 452.2258). − M.p. 136−138°C. − HPLC: R_t = 8.8 min (H_2O/CH_3CN/TFA, 4:1:0.1). − $[\alpha]_D^{25}$ = +10.2 (*c* = 1, MeOH).

4-Amino-4'-[N,N'-bis(tert-butyloxycarbonyl)guanidino]-benzophenone (**16**): 1060 mg of diaminobenzophenone (5 mmol) and 280 mg of *N,N'*-bis(*tert*-butyloxycarbonyl)thiourea (1 mmol) were dissolved in 5 ml of DMF. The solution was cooled to −10°C, and 0.6 ml of pyridine and 1.2 mmol of $HgCl_2$ were added. The reaction mixture was stirred for 2 h at −10°C and for 1 h at room temperature. Then, 20 ml of AcOEt was added and the resulting solution was filtered through Celite. The filtrate was washed with brine and then with 0.5 N HCl. The organic layer was dried over $MgSO_4$ and the solvent was evaporated. The oily residue was purified by column chromatography (dichloromethane/AcOEt, 4:1) yielding 290 mg (64%) of a yellow powder. − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.46 (s, 18 H), 4.21 (m, 2 H), 6.58 (d, 2 H, *J* = 8 Hz), 7.60 (d, 2 H, *J* = 8 Hz), 7.63 (m, 4 H), 10.45 (m, 1 H), 11.59 (m, 1 H). − MS (NBA): *m/z* = 455 ([M + H]$^+$). − M.p. 96−98°C. − R_f = 0.73 (dichloromethane/AcOEt, 4:1).

17: According to standard procedure A, compound **16** was coupled with **4** to afford **17** in 44% yield as a yellow oil after purification by column chromatography (dichloromethane/AcOEt, 4:1). − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.47 (s, 9 H), 1.49 (s, 9 H), 1.58−1.66 (m, 8 H), 2.25 (m, 1 H), 2.83−3.00 (m, 2 H), 3.56 (s, 3 H), 3.95 (d, 1 H, *J* = 16.5 Hz), 4.12 (m, 1 H), 4.36−4.51 (m, 2 H), 4.62 (m, 1 H), 7.58−7.70 (m, 8 H), 8.90 (m, 1 H), 10.59 (m, 1 H), 11.61 (m, 1 H). − MS (NBA): *m/z* = 761 ([M + H]$^+$). − R_f = 0.45 (dichloromethane/AcOEt, 4:1)

19: Deprotection of **17** followed by purification by HPLC afforded **19** as a yellow powder in 76% overall yield. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.64−1.95 (m, 8 H), 2.53 (m, 1 H), 2.88 (m, 1 H), 3.08 (m, 1 H), 4.10 (m, 1 H), 4.45 (m, 4 H), 7.44 (d, 2 H, *J* = 8 Hz), 7.73 (m, 3 H), 7.86 (m, 3 H). − ($C_{28}H_{31}N_6O_6$): HRMS = 547.2328 (for 547.2305). − M.p. 195−197°C. − HPLC: R_t = 10.1 min (H_2O/CH_3CN/TFA, 75:25:0.1). − $[\alpha]_D^{25}$ = + 8.5 (*c* = 1, MeOH).

N-(Fluorenylmethyloxycarbonyl)isonipecotic Acid **20**: 2 mmol of isonipecotic acid was dissolved in 14 ml of a 1:1 water/acetone mixture. The pH was adjusted to 8.5 with a saturated solution of $NaHCO_3$. A solution of 2 mmol of FmocOSu in 7 ml of acetone was added dropwise while the pH was kept constant by the addition of $NaHCO_3$. The reaction mixture was stirred for 3 h at room temperature and then 35 ml of AcOEt was added at 0°C. The resulting solution was acidified with $KHSO_4$ (1 M) and the aqueous layer was extracted with AcOEt. The combined organic phases were dried over $MgSO_4$ and the solvent was evaporated to afford 700 mg of a white powder (quantitative yield). − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.66−1.94 (m, 4 H), 2.53 (m, 1 H), 2.95 (m, 2 H), 4.11 (m, 2 H), 4.14 (m, 1 H), 4.43 (m, 2 H), 7.26−7.78 (m, 8 H). − MS (NBA): *m/z* = 352 ([M + H]$^+$). − M.p. 168−170°C. − R_f = 0.4 ($MeOH/NH_4OH$, 99:1).

23: According to general procedure A, **20** was coupled with the monoprotected diamines **5a**, **b** and **c** to afford **21a**, **b** and **c** in yields of 40, 47 and 50%, respectively. After removal of Boc in 80% yield, the deprotected compounds **22a**, **b** and **c** were coupled with **4** according to general procedure B to afford **23a**, **b** and **c** in respective yields of 53, 65 and 62%, based on **21a**, **b** and **c**.

23a: ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.52−1.92 (m, 12 H), 2.50 (m, 2 H), 2.70−2.96 (m, 4 H), 3.45−3.53 (m, 6 H), 3.70 (s, 3 H), 3.87 (d, 1 H, *J* = 16 Hz), 4.08−4.26 (m, 6 H), 4.60−4.71 (m, 3 H), 7.27−7.78 (m, 8 H). − MS (NBA): *m/z* = 700 ([M + H]$^+$). − R_f = 0.4 (AcOEt/MeOH, 8:2).

23b: ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.24 (m, 4 H), 1.38−1.62 (m, 10 H), 2.25 (m, 1 H), 2.57 (m, 1 H), 2.81 (m, 2 H), 3.06 (m, 2 H), 3.18 (m, 4 H), 3.67 (s, 3 H), 3.76 (d, 1 H, *J* = 19 Hz), 4.03−4.22 (m, 5 H), 4.40 (m, 4 H), 6.17 (m, 1 H), 6.90 (m, 1 H), 7.31−7.71 (m, 8 H). − MS (GT): *m/z* = 714 ([M + H]$^+$). − R_f = 0.52 (AcOEt/MeOH, 8:2).

23c: ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.18−1.34 (m, 4 H), 1.51−1.86 (m, 12 H), 2.25 (m, 1 H), 2.60 (m, 1 H), 2.69 (m, 2 H), 2.86 (m, 2 H), 3.05−3.25 (m, 4 H), 3.69 (s, 3 H), 3.86 (d, 1 H, *J* = 16 Hz), 4.13−4.40 (m, 6 H), 4.55 (m, 3 H), 6.36 (m, 1 H), 6.86 (m, 1 H). 7.21−7.65 (m, 8 H). − MS (NBA): *m/z* = 728 ([M + H]$^+$). − R_f = 0.68 (AcOEt/dichloromethane, 8:2).

Compounds **24**: 0.3 mmol of one of the above products **23a**, **b** or **c** was dissolved in 4 ml of MeOH and 0.7 ml of NaOH (1 N) was

added. The solution was stirred for 15 h at room temperature. The mixture was then neutralized with HCl (1 N) and the solvents were evaporated. The residue was purified by reversed-phase HPLC.

70 mg (53%) of **24a** was obtained as a white powder. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.54−2.04 (m, 12 H), 2.50 (m, 2 H), 2.84 (dd, 1 H, *J* = 12 Hz, *J* = 4 Hz), 2.95−3.06 (m, 3 H), 3.22−3.50 (m, 6 H), 4.04 (d, 1 H, *J* = 16 Hz), 4.27−4.36 (m, 4 H). − MS (GT): *m/z* = 464 ([M + H]$^+$). − HRMS ($C_{22}H_{34}N_5O_6$): 464.2536 (for 464.2509). − M.p. 198−200°C. − HPLC: R_t = 6.8 min (H_2O/CH_3CN, 9:1). − $[\alpha]_D^{25}$ = +10.2 (*c* = 1, MeOH).

90 mg (67%) of **24b** was obtained as a white powder after purification. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.56−1.74 (m, 6 H), 1.80−2.03 (m, 8 H), 2.50 (m, 1 H), 2.54 (m, 1 H), 2.62−2.72 (m, 2 H), 3.02 (td, 2 H, *J* = 12 Hz, *J* = 3 Hz), 3.22 (m, 4 H), 3.40 (m, 2 H), 4.18 (d, 1 H, *J* = 16 Hz), 4.24 (d, 1 H, *J* = 16 Hz), 4.36 (m, 3 H). − ($C_{23}H_{36}N_5O_6$): HRMS (GT): *m/z* = 478.2654 (for 478.2666). − M.p. 192−194°C. − HPLC: R_t = 6.3 min (H_2O/CH_3CN, 1:1). − $[\alpha]_D^{25}$ = +8.8 (*c* = 1, MeOH).

90 mg (65%) of **24c** was obtained as a white powder after purification. − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.47−1.92 (m, 16 H), 2.45 (m, 2 H), 2.93−3.15 (m, 6 H), 3.36−3.49 (m, 4 H), 4.00 (d, 1 H, *J* = 16 Hz), 4.22 (d, 1 H, *J* = 16 Hz), 4.27 (m, 3 H). − ($C_{24}H_{38}N_5O_6$): HRMS (NBA): *m/z* = 492.2820 (492.2822). − M.p. 90−92°C. HPLC: R_t = 12.1 min (H_2O/CH_3CN/TFA, 85:15:0.1). − $[\alpha]_D^{25}$ = +9.3 (*c* = 1, MeOH).

4-[N-(Benzyloxycarbonyl)aminoiminomethyl]aniline **25**: To a solution of 850 mg of aminobenzamidine dihydrochloride in 20 ml of THF and 4 ml of H_2O, was added 2.5 ml of 5 N NaOH. Then, 0.5 ml of benzyloxycarbonyl chloride in 13 ml of THF was slowly added dropwise at 0°C. The reaction mixture was stirred for 1 h at room temperature. The phases were then separated and the organic layer was concentrated to dryness. The residue was taken up in AcOEt and the resulting solution was washed with water. The organic layer was dried and the solvent was evaporated. The residue was crystallized from $CHCl_3$, affording 550 mg of **25** as a yellow powder (50% yield). − ^{1}H NMR ([D$_6$]DMSO, 250 MHz): δ = 5.03 (s, 2 H), 5.82 (s, 2 H), 6.51 (d, 2 H, *J* = 8 Hz), 7.33 (m, 5 H), 7.73 (d, 2 H, *J* = 8 Hz), 8.66 (m, 1 H), 9.11 (m, 1 H). − MS (NBA): *m/z* = 270 ([M + H]$^+$). − M.p. 140−142°C (ref.[12] 147−148°C). − HPLC: *R*t = 8.4 min (H_2O/CH_3CN/TFA, 85:15:0.1). − R_f = 0.55 (AcOEt/dichloromethane, 1:1).

26: 480 mg of Boc-Gly-OH (2.7 mmol) was dissolved in 10 ml of dichloromethane. At 0°C, 280 mg of DCC (1.35 mmol) was added. The reaction mixture was stirred for 1 h at 0°C and then filtered. The filtrate was added to a solution of 540 mg (2 mmol) of **25** in 10 ml of dichloromethane, and then 30 mg of DMAP and 0.3 ml of NEt_3 were added. The resulting mixture was stirred for 15 h at room temperature. The solvents were then evaporated, the residue was taken up in AcOEt, and the resulting solution was washed with 1 N HCl. The combined organic layers were dried and the solvent was evaporated. The residue was purified by column chromatography (AcOEt/dichloromethane, 1:1), affording 360 mg of a white powder (42% yield). − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.46 (s, 9 H), 3.88 (m, 2 H), 5.32 (s, 2 H), 5.98 (m, 1 H), 6.62 (m, 1 H), 7.30−7.58 (m, 9 H), 9.19 (m, 2 H). − MS (NBA): *m/z* = 427 ([M + H]$^+$). − M.p. 124−126°C. − R_f = 0.60 (AcOEt/dichloromethane, 1:1).

28: Compound **26** was deprotected (Boc removal) to afford **27** in 74% yield. According to general procedure A, 1.26 mmol of **4** and 1.26 mmol of **27** were coupled to give, after purification by column chromatography (AcOEt/dichloromethane, 1:1), **28** as a yellow powder in 31% yield. − ^{1}H NMR ($CDCl_3$, 250 MHz): δ = 1.51−1.86 (m, 8 H), 2.48 (m, 1 H), 2.94 (m, 2 H), 3.63 (s, 3 H), 3.68−3.79 (m, 4 H), 4.03 (m, 1 H), 4.37 (m, 2 H), 4.54 (m, 1 H), 5.18 (s, 2 H), 7.32−7.87 (m, 9 H), 8.23 (m, 1 H), 9.08 (m, 1 H), 9.47 (m, 1 H). − MS (NBA): *m/z* = 633 ([M + H]$^+$). − M.p. 132−134°C. − R_f = 0.4 (AcOEt/dichloromethane, 1:1).

30: Saponification and subsequent hydrogenolysis of 250 mg of **28** afforded 70 mg of **30** as a yellow powder after purification by HPLC (36%). − ^{1}H NMR (CD_3OD, 250 MHz): δ = 1.39−1.81 (m, 8 H), 2.47 (m, 1 H), 2.77 (dd, 1 H, *J* = 4 Hz, *J* = 16 Hz), 3.09 (dd, 1 H, *J* = 4 Hz, *J* = 16 Hz), 3.95 (d, 1 H, *J* = 16 Hz), 4.00 (m, 1 H), 4.12 (d, 1 H, *J* = 16 Hz), 4.32 (m, 4 H), 7.74 (d, 2 H, *J* = 9 Hz), 7.87 (d, 2 H, *J* = 9 Hz). − $C_{23}H_{39}N_6O_6$: HRMS (NBA): *m/z* = 485.2162 (485.2149). − M.p. 108−110°C. − HPLC: R_t = 11.1 min (H_2O/CH_3CN/TFA, 85:15:0.1). − R_f = 0.40 (MeOH). − $[\alpha]_D^{25}$ = +8.8 (*c* = 1, MeOH).

[1] C. Prasad, *Peptides* **1995**, *16*, 155.
[2] D. W. Gordon, J. Steele, *Bioorg. Med. Chem. Lett.* **1995**, *5*, 47.
[3] B. O. Scott, A. C. Siegmund, C. K. Marlowe, Y. Pei, K. L. Spear, *Molecular Diversity* **1995**, *1*, 125.
[4] N. Kucharczyk, C. Thurieau, J. Paladino, A. D. Morris, J. Bonnet, E. Canet, J. E. Krause, D. Regoli, R. Couture, J. L. Fauchère, *J. Med. Chem.* **1993**, *36*, 1654.
[5] For reviews on analogues and mimetics of RGD see: I. Ojima, Q. Dong, S. Chakravarty, E. Peerschke, S. M. Hwang, A. S. Wong, *Bioorg. Med. Chem.* **1995**, *5*, 1941; J. A. Zablocki, M. Miyano, N. R. Shashidhar, S. Panzer-Knodle, N. Nicholson, L. Feigen, *J. Med. Chem.* **1992**, *35*, 4914; V. Austel, F. Himmelsbach, T. Müller, *Drugs Fut*. **1994**, *19*, 757. For recent literature see: J. Lefkovits, E. Plow, E. J. Topol, *New Engl. J. Med.* **1995**, *332*, 1553.
[6] G. Marguerie, *French Patent* **1988**, *2*, 608, 160; G. Marguerie, *Chem. Abstr.* **1989**, *110*, 115347a; F. S. Tjoeng, S. P. Adams, *U.S. Patent* **1989**, 4,879,313.
[7] G. D. Hartman, M. S. Egbertson, W. Halczenko, W. L. Laswell, M. E. Duggan, R. L. Smith, A. M. Naylor, P. D. Manno, R. J. Lynch, G. Zhang, C. T. C. Chang, R. J. Gould, *J. Med. Chem.* **1992**, *35*, 4640; M. E. Duggan, A. M. Naylor-Olsen, J. J. Perkins, P. S. Anderson, C. T. C. Chang, J. J. Cook, R. J. Gould, N. C. Ihle, G. D. Hartman, J. J. Lynch, R. J. Lynch, P. D. Manno, L. W. Schaffer, R. L. Smith, *J. Med. Chem.* **1995**, *38*, 3332.
[8] J. A. Zablocki, M. Miyano, R. B. Garland, D. Pireh, L. Schretzman, N. R. Shashidhar, S. N. Rao, R. J. Lindmark, S. Panzer-Knodle, N. Nicholson, B. B. Taite, A. K. Salyers, L. W. King, J. G. Campion, L. P. Feigen, *J. Med. Chem.* **1993**, *36*, 1811.
[9] I. Ojima, Q. Dong, M. Eguchi, Y. I. Oh, C. M. Amman, B. S. Coller, *Bioorg. Med. Chem. Lett.* **1994**, *4*, 1749.
[10] D. Ben-Ishai, E. Goldstein, *Tetrahedron* **1971**, *27*, 3119; M. Vincent, C. Pascard, M. Cesario, G. Rémond, J. P. Bouchet, Y. Charton, M. Laubie, *Tetrahedron Lett.* **1992**, *33*, 7369.
[11] K. S. Kim, L. Qian, *Tetrahedron Lett.* **1993**, *34*, 7677; E. J. Iwanowicz, M. A. Poss, J. Lin, *Synth. Commun.* **1993**, *23*, 1443.
[12] W. E. Bondinell, R. M. Keenan, W. H. Miller, F. E. Ali, A. C. Allen, C. W. De Brosse, D. S. Eggleston, K. F. Erhard, R. C. Haltiwanger, W. F. Huffman, S. M. Hwang, D. R. Jakas, P. F. Koster, T. W. Ku, C. P. Lee, A. J. Nichols, S. T. Ross, J. M. Samanen, R. E. Valocik, J. A. Vasko-Moser, J. W. Venslavsky, A. S. Wong, C. K. Yuan, *Bioorg. Med. Chem.* **1994**, *2*, 897.
[13] M. B. Zucker, *Meth. Enzymol.* **1988**, *169*, 117.
[14] V. Austel, W. G. Eiser, F. Himmelsbach, G. Linz, T. H. Miller, H. Pieper, E. Seewaldt-Becker, H. Weisenberger, *207th American Chemical Society National Meeting*, MEDI, **1993**, 101.
[15] P. Martin, T. Papayamopoulou, *Science* **1982**, *216*, 1233.
[16] A. Molla, R. Berthier, A. Chapel, A. Schweitzer, A. Andrieux, *Biochem. J.* **1995**, *309*, 491.

[97320]