β-Lactam-Containing Cyclopeptide Analogs

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Dedicated to János Rétey on the occasion of his 70th birthday

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Cyclic peptide analogs containing a β-lactam moiety were prepared. Reacting Fmoc-protected amino acid-derived diazo ketones **1**, **2** with benzylidene-protected amino esters **3**, **4** in a photochemically induced Staudinger-type reaction, *trans*-substituted β-lactams **5a/b** and **6a/b** were formed in 35–70% yield (*dr* 60:40–70:30). *N*-Terminal chain elongation to the respective acyclic precursors **14**, **17a/b** and **19a/b** was achieved using conventional peptide synthesis (i.e. the pentafluorophenyl ester protocol). After saponification, activation as pentafluorophenyl esters and subsequent cleavage of the *N*-terminal Boc-protecting group, the pre-strained (3*R*,4*S*)-configured isomers could be cyclized without the need for high dilution or prolonged reaction times. Contrary to this, the (3*S*,4*R*)-configured isomers did not cyclize but gave polymeric material. The conformational stability of the cyclic peptidomimetics **16**, **20**, and **21** which were obtained in high yield, was elucidated by means of NMR spectroscopy.

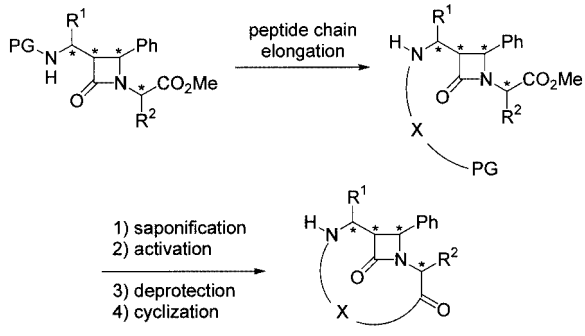
Introduction

Peptides showing a turn conformation[1–4] are of major relevance since, in most cases, a turn region is responsible for the biological activity of the respective peptide. Well known representatives showing a turn conformation are, inter alia, somatostatin[5–7] and oxytocin.[8] Replacing the peptide turn by other, mostly cyclic templates[9–11] furnishes substrates with a more rigid framework, that are potentially capable of mimicking a biologically active conformation (i.e., turn mimics). Many peptidic hormones have a cyclic structure resulting in further advantageous features. Not only is a possible biologically active conformation more rigidly fixed, cyclic substrates are occasionally less polar which makes separation and purification easier and they are usually more stable against digesting enzymes what allows for an oral application. Nevertheless, the cyclization step in the synthesis of cyclic peptides is often limiting the value of the method and low yields have to be taken into account.

During our investigations on peptidomimetics containing β-lactams we found that these give very stable turn structures.[12] Crystal structure analysis showed that the angle between incoming and leaving peptide strands (i.e. the vectors at which the peptide chain enters and leaves the turn mimetic) is about 185°. We envisioned that this pre-strained turn conformation might lead to an efficient cyclization of appropriately prepared precursors. In this paper we report on our efforts in this context.

Results and Discussion

For the preparation of the β-lactam-containing peptide fragments we used a method developed in our group in which α-amino acid-derived diazo ketones were photochemically reacted with imines derived from α-amino esters (photochemically induced Staudinger-type reaction). Although this reaction is similarly possible with larger, peptide-derived substrates,[13] we abolished this approach, since yields were significantly lower therein. Instead, we used conventional peptide coupling procedures for the elongation of the corresponding amino acid-derived β-lactams (Scheme 1).



Scheme 1. General scheme for the preparation of β-lactam-containing cyclopeptide analogs

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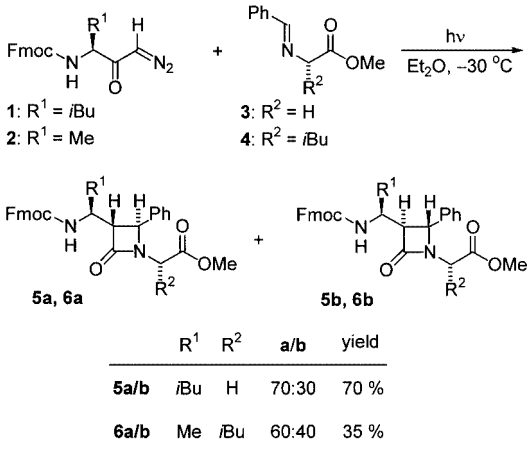
The diazo ketones utilized in this protocol were prepared according to published procedures by reacting carbamate-protected amino acids (activated via the mixed anhydride method) with diazomethane.[14] Synthesis of the imine compounds was achieved by liberation of amino esters from their hydrochlorides with ammonia and subsequent condensation with benzaldehyde in the presence of alumina.[15,16]

Fmoc-protected diazo ketones **1** and **2** derived from leucine and alanine, respectively, were treated with *N*-(benzylidene)glycine and leucine methyl ester, respectively (**3**, **4**), and mixtures of the corresponding diastereomeric *trans*-substituted β-lactams were obtained (Scheme 2). Selectivities were in accordance with prior observations, depending on the bulkiness of the diazo ketones' side-chains. Alanine-derived substrates gave a 60:40 selectivity (**5a/b**), while leucine-derived diazo ketones led to a slightly better 70:30 selectivity (**6a/b**). The isomers were separated by means of medium pressure liquid chromatography (MPLC).



	R^1	R^2	a/b	yield
5a/b	*i*Bu	H	70:30	70 %
6a/b	Me	*i*Bu	60:40	35 %

Scheme 2. Synthesis of β-lactam-containing dipeptide analogs

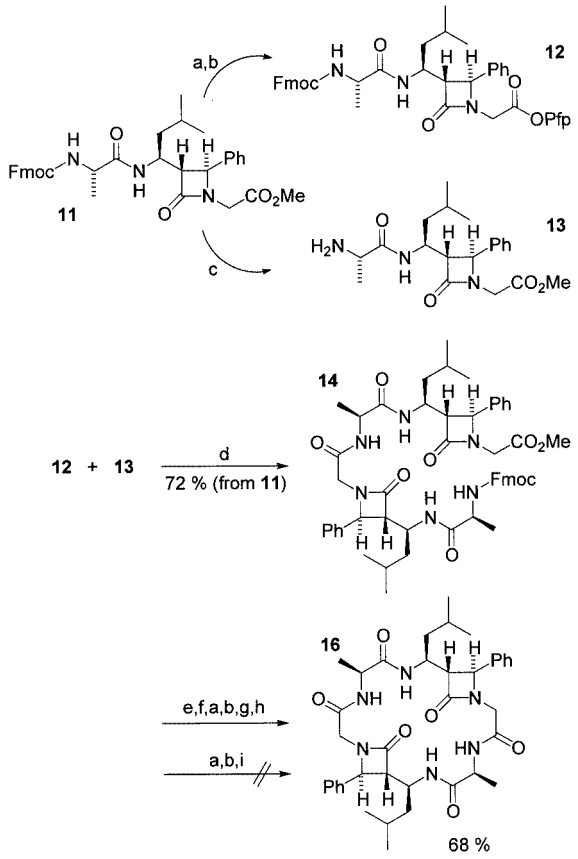
As a first cyclic target molecule we chose a symmetric analog containing two β-lactam moieties. In this case we expected a cyclization to be especially favored. In addition, spectroscopic analysis should be simplified due to the C_2 symmetry of the substrate and − if the region containing the β-lactam is responsible for a possible biological activity − a doubling of this region might lead to an increased effect.

Synthesis of the precursor for the cyclization step was straightforward. (3*R*,4*S*)-Configured azetidinone **5a** was deprotected at the *N*-terminus with diethylamine, a secondary base which was easily removed in vacuo when the reaction was complete. The thus liberated amino compound was immediately reacted with Fmoc−Ala−OPfp (**8**) resulting in the formation of tripeptide analog **11**. Half the material was saponified with lithium hydroxide with no cleavage of the base-labile Fmoc group[17] and was subsequently activated as pentafluorophenyl ester [prepared from *N*-ethyl-*N*′-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC) and pentafluorophenol (PfpOH)]. The remaining fraction of intermediate **11** was deprotected at the *N*-terminus with tris(2-aminoethyl)amine (TAEA)[18] and added without further purification to the priorly synthesized active ester furnishing the linear precursor **14** in 72% yield.

Saponification of **14** was achieved by employing drastically prolonged reaction times (ca. 40 h) which led to a partial cleavage of the Fmoc group. The pentafluorophenyl ester protocol was again used for activation of the carboxylic acid. However, when the Fmoc-protected amino function was deprotected under basic conditions (1,8-diazabicyclo[5.4.0]undec-7-ene, DBU, or 1,5-diazabicyclo[4.3.0]non-5-ene, DBN, respectively) to achieve cyclization, cleavage of the pentafluorophenyl ester occurred; no cyclized product could be isolated. Instead of investigating further modifications of the reaction protocol we decided to use an optimized procedure developed for the cyclization of peptides by Schmidt et al.[19,20] For this purpose we had to exchange the Fmoc group for the Boc group (→ **15**), which was achieved by deprotection with diethylamine and subsequent treatment of the liberated amine with di-*tert*-butyl dicarbonate (Boc_2O). Saponification was now − due to the base stability of the Boc group − possible without any loss and activation was effected as described above. Further purification of the obtained pentafluorophenyl ester was not necessary since NMR spectroscopy showed that the material was essentially pure. Cleavage of the Boc group with trifluoroacetic acid (TFA) prior to slow addition to an emulsion of chloroform and 1 M potassium carbonate solution without the need for high dilution gave the cyclized peptidomimetic **16** in 68% yield (from **14**) within 2 h (Scheme 3).

The pre-formed turn structure of the β-lactam-containing precursors should also facilitate the often poor yielding cyclization of smaller cyclopeptide analogs built from four and five amino acid residues, respectively. Attempts to synthesize such compounds in a similar way as outlined above were actually successful: The acyclic precursors were prepared from dipetide analogs **5a/b** by successive attachment of two amino acid residues at the *N*-terminus and from dipetide analogs **6a/b** by coupling with a Boc-protected tripeptide. All couplings were accomplished using the pentafluorophenyl ester method (Scheme 4). Yields were rather poor for the segment coupling furnishing **19a/b** since significant amounts of the epimerized product (at C-1 of Phe) had to be separated. This epimerization possibly occurred prior to the activation of Boc−Ala−Ala−Phe−OH during saponification of the corresponding methyl ester **18**,[4] or during activation of Boc−Ala−Ala−Phe−OH, though we never did encounter an epimerization during pentafluorophenyl ester activation in other cases. Given yields refer to analytically and diastereomerically pure material.

While saponification of the tetrapeptide analog **17a** was achieved with aqueous lithium hydroxide within two hours, the cleavage of the other methyl esters was complete only after drastically prolonged reaction times (> 20 h for **17b** and **19b**) or when sodium hydroxide in dimethylformamide was used (18 h for **19a**). However, the stereochemical integrity of the material was maintained even under these con-

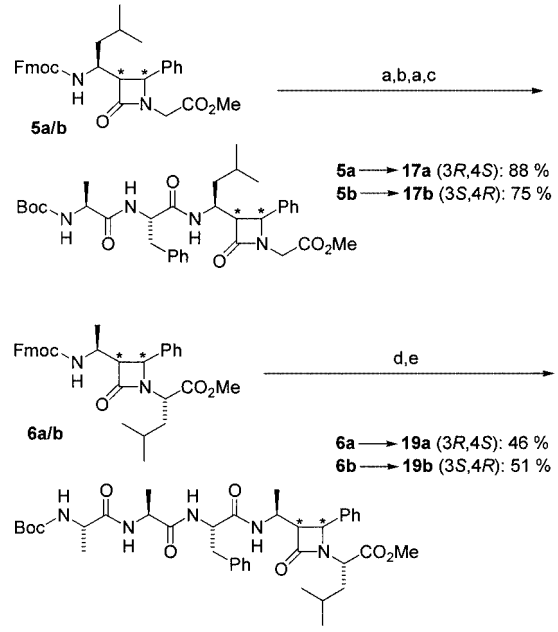


Scheme 3. Synthesis of cyclic peptidomimetic **16**. a) 0.2 N $LiOH_{aq.}$, THF, 0 °C; b) PfpOH, EDC, CH_2Cl_2, 0 °C to room temp.; c) TAEA, CH_2Cl_2, room temp.; d) CH_2Cl_2, room temp., 72%; e) Et_2NH, CH_2Cl_2, room temp.; f) Boc_2O, CH_2Cl_2, room temp., 91%; g) TFA, 0 °C; h) 1 M $K_2CO_{3\,aq.}$, $CHCl_3/CH_2Cl_2$, room temp., 75%; i) DBU or DBN, THF or CH_2Cl_2, room temp.

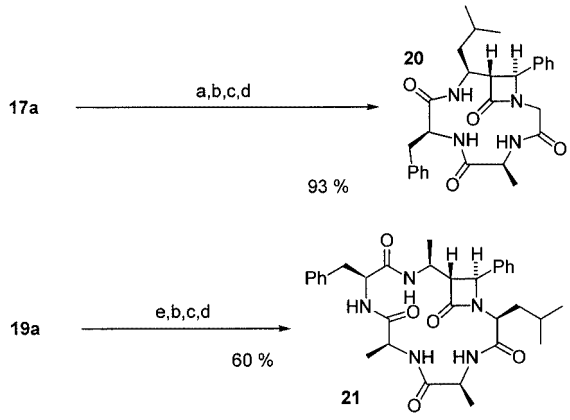
ditions. Cyclization of the (3*R*,4*S*)-configured carboxylic acids obtained from **17a** and **19a**, respectively, was achieved under the above described conditions, yielding peptide analogs **20** and **21**, respectively, in notably high yields (93 and 60%, Scheme 5).

On the other hand, the (3*S*,4*R*)-configured precursors **17b** and **19b** could not be cyclized, even under high dilution conditions ($\approx 2\cdot10^{-3}$ M) and with prolonged reaction times (up to 10 h). In lieu thereof oligomeric material was obtained which − due to its poor solubility − was not further purified. The reason for this differing observation is not fully understood. Some evidence comes from simple modelling of the respective cycles. All considered conformations (with different starting geometries) proved to be significantly more stable with a (3*R*,4*S*) configuration. Though the cause for this is not clear (steric reasons should be responsible), it might similarly count for the deviating tendencies for cyclization. However, a complete survey of the conformational space, which would give more evidence, was not performed.

Due to the inherent symmetry of analog **16**, its NMR spectra are very concise. The main structural feature of this compound is − apart from the β-lactam − a hydrogen bond between the β-lactam carbonyl and the amide hydrogen of



Scheme 4. Synthesis of acylic precursors **17a/b** and **19a/b**. a) Et_2NH, THF, room temp.; b) Fmoc−Phe−OPfp (**7**), THF, room temp.; c) Boc−Ala−OPfp (**9**), THF, room temp.; d) Et_2NH, MeCN, room temp.; e) Boc−Ala−Ala−Phe−OPfp, THF, room temp.



Scheme 5. Synthesis of cylic peptidomimetics **20** and **21** containing one β-lactam moiety. a) 0.2 N $LiOH_{aq.}$, THF, 0 °C; b) PfpOH, EDC, CH_2Cl_2, 0 °C to room temp.; c) TFA, 0 °C; d) 1 M $K_2CO_{3\,aq.}$, $CHCl_3/CH_2Cl_2$, room temp.; e) 1 M $NaOH_{aq.}$, DMF, room temp.

the neighboring leucine that forms a six-membered ring attached to the azetidinone moiety. Evidence for this structural motif, which has already been recognized in the corresponding acyclic peptidomimetics,[12] comes from a downfield shift of the amide proton and the coupling constants (allowing for an estimation of the respective angles) of the protons in this region (see Supporting Information for further information, see also the footnote on the first page of this article). The geminal coupling of the glycine α-protons is rater high (16 Hz) suggesting a bisectal conformation. Chemical shifts of most protons are hardly dependent on the concentration. However, slight changes of chemical shifts for the amide protons and a significant increase of the distance between the signals of the glycin α-

Table 1. Chemical shifts of selected protons in cyclic peptidomimetic **16** and their concentration dependancy

Concentration	$NH_{leucine}$	$NH_{alanine}$	α-$H_{alanine}$	H-$4_{\beta\text{-lactam}}$	α-$H_{leucine}$	α_a-$H_{glycine}$	α_b-$H_{glycine}$	H-$3_{\beta\text{-lactam}}$
0.001 M	7.68	6.67	4.53	4.50	4.49	3.87	3.77	3.17
0.050 M	7.64	6.77	4.53	4.51	4.50	3.89	3.74	3.16
0.180 M	7.58	6.99	4.53	4.53	4.49	3.96	3.66	3.15

protons with increasing concentration show a change in the molecule's geometry, most likely due to some sort of intermolecular interaction. The signal of the leucine amide proton forming the hydrogen bond shows almost no solvent or concentration dependency [$\Delta\delta$(leucine-NH) = +0.05 ppm for a change from $CDCl_3$ to $CDCl_3/CD_3OD$ (5:1) as compared with $\Delta\delta$(alanine-NH) = +0.47 ppm for $CDCl_3$ to $CDCl_3/CD_3OD$ (5:1)] (Table 1). However, these findings are in disagreement with a fast H/D exchange of this proton [leucine-NH: about 5 min when changing from $CDCl_3$ to $CDCl_3/CD_3OD$ (5:1) as compared with alanine-NH: about 30 min when the solvent was changed from $CDCl_3$ to $CDCl_3/CD_3OD$ (5:1)].

Signals of the cyclic tetrapeptide analog **20** are significantly broadened at room temperature giving evidence for a more floppy structure. Sharper signals revealing the coupling pattern were obtained only at 330 K. Although the signal of the amide proton next to the β-lactam carbonyl in this peptide analog also appears at significantly lower field compared to the other amide protons, the hydrogen bond (with a participation of the β-lactam carbonyl) seems − most probably due to steric reasons − to be not present or at least rather weak in this substrate. The conformation of cyclopeptide analog **21** seems to be very sensitive towards the conditions under which the spectra are recorded. While the NMR spectra in CD_2Cl_2 show sharp signals even at room temperature, only broadened signals could be observed when $CDCl_3$ was used as a solvent. The ambiguous data obtained for this substrate may be explained by coordination of different amounts of water to the cyclopeptide mimetic since traces of water were unavoidable in this case.

Conclusion

We were able to show that a (3*R*,4*S*)-configured β-lactam moiety incorporated into small peptides enforces a turn conformation leading to a facilitated cyclization of suitable precursors. This is not only valid for larger rings but as well for small cyclopetide analogs consisting of only four or five amino acid residues, respectively. Contrary to this, the (3*S*,4*R*)-configured isomers could not be cyclized but gave polymeric material.

Experimental Section

General Remarks: Solvents for chromatography and for workup, e.g. ethyl acetate and light petroleum (PE) were distilled prior to use, diethyl ether was distilled from $KOH/FeSO_4$. Diethyl ether and THF used for reactions were distilled from sodium/benzophenone. Diazo ketones **1** and **2**[14] were prepared as described elsewhere. Fmoc- and Boc-protected amino acids and amino ester hydrochlorides were purchased from Bachem, Boc−Ala−Ala−Phe−OMe (**18**) and pentafluorphenyl esters **7**−**9** were purchased from Bachem or prepared according to standard protocols.[4] Common amino acid abbreviations are used.[21] EDC: *N*-ethyl-*N*′-[3-(dimethylamino)propyl]carbodiimide hydrochloride, TAEA: tris(2-aminoethyl)amine. Flash column chromatography: Merck silica gel 60 (230−400 mesh). TLC: precoated sheets from Macherey−Nagel, Alugram SIL G/UV_{254}; detection by UV extinction, by cerium molybdatophosphate solution [phosphomolybdic acid (25 g), $Ce(SO_4)_2 \cdot H_2O$ (10 g), concd. H_2SO_4 (60 mL), H_2O (940 mL)], by anisaldehyde solution [anisaldehyde (9.2 mL), AcOH (3.75 mL), H_2SO_4 (12.5 mL), EtOH (335 mL)] or by ninhydrin solution (2% in EtOH). Medium pressure liquid chromatography (MPLC): Detection by UV absorption (Latek UVIS 200). HPLC: Analyses of diastereoisomer distribution were carried out with a Pharmacia LKB, RSD 2140 apparatus with a Pharmacia LKB, RSD 2249 mixer and diode-array detection (Pharmacia RSD 2140) with a Merck LiChrosorb Si 60 chromatographic column (hexane/EtOAc, flow: 2.0 mL/min). ^1H and ^{13}C NMR spectra were recorded with a Bruker ARX 500 spectrometer at room temp. in $CDCl_3$ unless otherwise indicated. Chemical shifts (δ in ppm) relative to internal TMS (0 ppm) or to resonances of the solvent (^1H: $CHCl_3$, 7.24 ppm; ^{13}C: $CDCl_3$, 77.0 ppm), coupling constants *J* in Hz. The numbering used for the assignment of the NMR spectroscopic data for the cyclic peptidomimetics **16**, **20**, and **21**; detailed spectroscopic data for all compounds and NMR spectra for key substances are included in the Supporting Information. Mass spectra were recorded using a Finnigan MAT 95 [FAB or CI (CH_4 or NH_3) technique] or a Varian MAT 711 instrument (EI). IR spectra were recorded with a Bruker IFS 28 or a Perkin−Elmer 283 instrument. Elemental analyses were performed by the service of the Institut für Organische Chemie, Stuttgart. Melting points are not corrected.

General Procedure for the Preparation of Imines 3 and 4:[15,16] The amino acid methyl ester hydrochloride (20.0 mmol) was suspended at 0 °C in a ca. 0.5 M solution of NH_3 in $CHCl_3$ (100 mL), stirred for 30 min, left without stirring for further 20 min, filtered (Celite®) and carefully concentrated in vacuo (300 mbar, 35 °C). Al_2O_3 (neutral, type 90, mesh 63−200 μm, activity grade I, 10.0 g), benzaldehyde (18.0 mmol) and CH_2Cl_2 (2 mL) were added, and the mixture was stirred at room temp. for 3 h. The suspension was filtered (Celite®) and washed with CH_2Cl_2. The solvent was evaporated (300 mbar, 35 °C) and the residue purified by a bulb-to-bulb distillation.

Dipeptide Analogs 5a,b: A quartz photo reactor was charged with Fmoc−Leu−CHN_2 (**1**, 2.72 g, 7.20 mmol), Bn=Gly−OMe (**3**, 2.04 g, 11.5 mmol), and Et_2O (280 mL), then flushed with N_2, cooled to −30 °C, and irradiated with a UV lamp for 1.5 h (monitoring by TLC) with vigorous stirring. The solution was warmed to room temp. and the solvent was removed (300 mbar, 35 °C). Excess imine was removed by flash chromatography (SiO_2). The

diastereomeric ratio was determined by HPLC (hexane/*i*PrOH, 95:5) and ^{1}H NMR spectroscopy (**5a/b** = 70:30). Separation of side products and impurities by MPLC (PE/EtOAc, 80:20) and separation of the isomers by MPLC (PE/*i*PrOH, 98:2) afforded the azetidinone **5a** (1.59 g, 3.02 mmol, 42%) and the azetidinone **5b** (0.76 g, 1.44 mmol, 20%) as colorless solids. A mixture of isomers was obtained in an additional fraction (303 mg, 576 μmol, 8%). **Major Isomer 5a:** M.p. 64−65 °C. $[\alpha]_D^{20}$ = −17.0 (*c* = 1.00, $CHCl_3$). $C_{32}H_{34}N_2O_5$ (526.62): calcd. C 72.98, H 6.51, N 5.32; found C 72.66, H 6.75, N 5.11. **Minor Isomer 5b:** M.p. 85−88 °C. $[\alpha]_D^{20}$ = +20.5 (*c* = 1.00, $CHCl_3$). $C_{32}H_{34}N_2O_5$ (526.62): calcd. C 72.98, H 6.51, N 5.32; found C 72.90, H 6.63, N 5.22.

Dipeptide Analogs 6a,b: Fmoc−Ala−CHN_2 (**2**, 2.08 g, 6.20 mmol) and Bn=Leu−OMe (**4**, 1.88 g, 8.06 mmol) in Et_2O (280 mL) were reacted as described for compounds **5a/b**. The diastereomeric ratio was determined by HPLC (hexane/EtOAc, 75:25) and ^{1}H NMR spectroscopy (**6a/b** = 60:40). Separation of side products and impurities by MPLC (PE/EtOAc, 88:12) and separation of the isomers by MPLC (PE/*i*PrOH, 98:2) afforded azetidinone **6a** (403 mg, 930 μmol, 15%) and azetidinone **6b** (369 mg, 682 μmol, 11%) as colorless foams. A mixture of isomers was obtained in an additional fraction (302 mg, 558 μmol, 9%). **Major Isomer 6a:** M.p. 63−71 °C (softening range). $[\alpha]_D$ ^{20} = −23.1 (*c* = 1.00, $CHCl_3$). $C_{33}H_{36}N_2O_5$ (540.65): calcd. C 73.31, H 6.71, N 5.18; found C 73.22, H 6.96, N 5.12. **Minor Isomer 6b:** M.p. 66−70 °C (softening range). $[\alpha]_D^{20}$ = +8.8 (*c* = 1.01, $CHCl_3$). $C_{33}H_{36}N_2O_5$ (540.65): calcd. C 73.31, H 6.71, N 5.18; found C 73.01, H 6.73, N 5.05.

Tripeptide Anolog 10a: A mixture of the azetidinone **5a** (300 mg, 570 μmol) and Et_2NH (753 mg, 1.07 mL, 10.3 mmol) in THF (3 mL) was stirred for ca. 5 h (monitoring by TLC). The volatile components were removed (20 mbar, 40 °C) and the residue was repeatedly dissolved in THF and the solvents were evaporated in order to remove traces of Et_2NH. The residue was dissolved in THF (6 mL) and Fmoc−Phe−OPfp (**7**, 376 mg, 680 μmol) was added. After stirring for 2.5 h the solvent was removed (100 mbar, 40 °C), the residue was dissolved in EtOAc, extracted with brine (3×), dried ($MgSO_4$), filtered, and concentrated in vacuo. Column chromatography (SiO_2, PE/EtOAc, 4:1 to 3:1) yielded the tripeptide analog **10a** (353 mg, 520 μmol, 92%) as a colorless foam. M.p. 89−91 °C (softening range). $[\alpha]_D^{20}$ = −25.6 (*c* = 0.99, $CHCl_3$). $C_{41}H_{43}N_3O_6$ (673.80): calcd. C 73.08, H 6.43, N 6.24; found C 72.55, H 6.45, N 6.15. HR-MS: calcd. for $^{12}C_{41}{}^{1}H_{44}{}^{14}N_3{}^{16}O_6$ 674.3241; found *m*/*z* = 674.3230.

Tripeptide Anolog 10b: The azetidinone **5b** (526 mg, 1.00 mmol) was deprotected with Et_2NH (1.83 g, 2.60 mL, 25.0 mmol) in THF (6 mL) and subsequently reacted with Fmoc−Phe−OPfp (**7**, 664 mg, 1.20 mmol) in THF (11 mL) as described for compound **10a**. Column chromatography (SiO_2, PE/EtOAc, 3:1) yielded the tripeptide analog **10b** (532 mg, 790 μmol, 79%) as a colorless foam. M.p. 96−99 °C (softening range). $[\alpha]_D^{20}$ = −8.5 (*c* = 1.00, $CHCl_3$). $C_{41}H_{43}N_3O_6$ (673.80): calcd. C 73.08, H 6.43, N 6.24 found C 72.74, H 6.48, N 6.13.

Tripeptide Anolog 11: The azetidinone **5a** (1.07 g, 2.03 mmol) was deprotected with Et_2NH (2.68 g, 3.81 mL, 36.6 mmol) in THF (17 mL) and subsequently reacted with Fmoc−Ala−OPfp (**8**, 1.16 g, 2.44 mmol) in THF (17 mL) as described for compound **10a**. Column chromatography (SiO_2, PE/EtOAc, 3:1) yielded the tripeptide analog **11** (1.14 g, 1.91 mmol, 94%) as a colorless foam. M.p. 94−96 °C (softening range). $[\alpha]_D^{20}$ = −49.4 (*c* = 1.01, $CHCl_3$). $C_{35}H_{39}N_3O_6$ (597.70): calcd. C 70.33, H 6.58, N 7.03; found C 70.36, H 6.41, N 6.85.

Fmoc-Protected Acyclic Precursor 14: To a solution of the tripeptide analog **11** (550 mg, 920 μmol) in THF (20 mL), 0.2 N aqueous LiOH (28 mL) was added in 3 portions at 0 °C within 1.5 h. Stirring was continued for 2 h, and the solution was added to a mixture of 0.2 N HCl (300 mL) and EtOAc (100 mL). The aqueous phase was extracted with EtOAc (2×) and the combined organic layers were dried ($MgSO_4$) and concentrated in vacuo (50 mbar, 40 °C). The residue was dissolved in CH_2Cl_2 (15 mL), the solution stirred and cooled to 0 °C, then pentafluorophenol (170 mg, 920 μmol) and EDC (194 mg, 1.01 mmol) were added. Stirring was continued at 0 °C for 2 h and at room temp. for 2 h. The solvents were removed (300 mbar, 35 °C), EtOAc and brine were added to the residue. The organic layer was extracted with saturated $NaHCO_3$ (2×) and with brine, dried ($MgSO_4$), filtered through a pad of Celite®, and the solvents evaporated (50 mbar, 40 °C) to yield the intermediate pentafluorophenyl ester **12**.

To a second portion of **11** (530 mg, 890 μmol) in CH_2Cl_2 (20 mL) TAEA (6.48 g, 6.63 mol, 44.3 mmol) was added, and the mixture was stirred for 10 min (monitoring by TLC), diluted with additional CH_2Cl_2, extracted with brine (2×), phosphate buffer (3×, pH 5.5: 6.54 g Na_2HPO_4 and 20.3 g NaH_2PO_4·2 H_2O in 100 mL H_2O), brine (1×), dried ($MgSO_4$), and concentrated in vacuo (100 mbar, 40 °C). The thus liberated amine **13** was diluted in CH_2Cl_2 (25 mL) and the pentafluorphenyl ester **12** was added. After stirring at room temp. for 2.5 h the solvents were removed (100 mbar, 40 °C), the residue was dissolved in EtOAc, extracted with brine (3×), dried ($MgSO_4$), filtered, concentrated in vacuo, and purified by column chromatography (SiO_2, PE/EtOAc, 1:2) to yield the Fmoc-protected acyclic precursor **14** (623 mg, 660 μmol, 74%) as a colorless foam. M.p. 120−123 °C. $[\alpha]_D^{20}$ = −52.6 (*c* = 1.01, $CHCl_3$). $C_{54}H_{64}N_6O_9$ (941.12): calcd. C 68.92, H 6.85, N 8.93; found C 68.58, H 6.87, N 8.85. HR-MS: calcd. for $^{12}C_{54}{}^{1}H_{65}{}^{14}N_6{}^{16}O_9$ 941.4813; found *m*/*z* = 941.4807.

Boc-Protected Acyclic Precursor 15: Et_2NH (1.28 g, 1.82 mL, 17.5 mmol) was added to a solution of the Fmoc-protected acyclic precursor **14** (551 mg, 580 μmol) in CH_2Cl_2 (10 mL) and the mixture was stirred at room temp. for 5 h (monitoring by TLC). The volatile components were removed (20 mbar, 40 °C) and the residue was repeatedly dissolved in THF and the solvents were evaporated in order to remove traces of Et_2NH. The residue was dissolved in CH_2Cl_2 (15 mL), Boc_2O (317 mg, 1.45 mmol) was added, and the solution was stirred at room temp. for 14 h. EtOAc was added and the mixture was extracted with saturated NH_4Cl solution (2×), saturated $NaHCO_3$ solution, and with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo (50 mbar, 40 °C). Column chromatography (SiO_2, PE/EtOAc, 2:3) afforded the Boc-protected acyclic precursor **15** (431 mg, 530 μmol, 91%) as a colorless foam. M.p. 118−121 °C (softening range). $[\alpha]_D^{20}$ = −69.6 (*c* = 1.00, $CHCl_3$). $C_{44}H_{62}N_6O_9$ (819.00): calcd. C 64.53, H 7.63, N 10.26; found C 64.65, H 7.77, N 9.99.

Cyclic Peptidomimetic 16: To a solution of Boc-protected acyclic precursor **15** (324 mg, 390 μmol) in THF (28 mL), 0.2 N aqueous LiOH (18 mL) was added in 3 portions at 0 °C within 1.5 h. Stirring was continued for 2 h and the solution was added to a mixture of 0.2 N HCl (300 mL) and EtOAc (100 mL). The aqueous phase was extracted with EtOAc (2×) and the combined organic layers were dried ($MgSO_4$) and concentrated in vacuo (50 mbar, 40 °C). The residue was dissolved in CH_2Cl_2 (13 mL), the solution stirred and cooled to 0 °C, then pentafluorophenol (72 mg, 0.39 mmol) and EDC (82 mg, 0.42 mmol) were added. Stirring was continued at 0 °C for 2 h and at room temp for another 2 h. The solvents were removed (300 mbar, 35 °C) and EtOAc and brine were added

to the residue. The organic layer was extracted with saturated $NaHCO_3$ (2×) and with brine, dried ($MgSO_4$), filtered through a pad of Celite®, and concentrated in vacuo (50 mbar, 40 °C).

Deprotection was achieved by treatment of the residue with TFA (4.47 g, 3.00 mL, 39.2 mmol) at 0 °C for 2.5 h. After removal of excess TFA in vacuo (20 mbar, 40 °C) the obtained salt was dissolved in CH_2Cl_2. The cyclization reaction was started by dropwise addition (1 h) of the CH_2Cl_2 solution to a vigorously stirred emulsion of 1 M aqueous K_2CO_3 (20 mL) and $CHCl_3$ (40 mL). The aqueous phase was separated after further stirring for 1 h and extracted with $CHCl_3$ (3×). The combined organic layers were dried ($MgSO_4$), filtered, and the solvent was evaporated (50 mbar, 40 °C). Column chromatography (SiO_2, PE/EtOAc, 1:1 to 1:3) yielded the cyclic peptidomimetic **16** (201 mg, 0.29 mmol, 75%) as a colorless solid. M.p. 164−166 °C. $[\alpha]_D^{20} = -74.8$ ($c = 1.03$, $CHCl_3$). IR (KBr): $\tilde{\nu}$ = 3395, 3290 (N−H), 3041, 3011 (arom. C−H), 2933, 2911, 2844 (aliphat. C−H), 1740, 1652 (C=O), 1520 (δ C−N−H + ν C−N, amide II), 1443 (δ C−H and C=C), 728, 677 (arom. C−H $_{o,o,p}$) cm^{-1}. MS (FAB, pos., matrix: NBA): *m/z* = 709 (23) $[M + Na]^+$, 687 (100) $[M + H]^+$, 189 (29) $[PhCH{=}NHCH_2C(O)NC_2H_4]^+$, 131 (25) $[PhC_3H_2O]^+$, 86 (26) $[H_2N{=}CHCH_2CHMe_2]^+$. ^1H NMR (500 MHz, $CDCl_3$): δ = 0.87, 0.92 (2 × d, ^3J = 6.6 Hz, 2 × 6 H, H_{Leu}-δ), 1.40 (ddd, ^2J = 13.9, ^3J = 8.9, ^3J = 5.1 Hz, 2 H, H_{Leu}-β_a), 1.53 (d, ^3J = 7.3 Hz, 6 H, H_{Ala}-β), 1.66 (dseptd, ^3J = 8.9, ^3J = 6.6, ^3J = 5.2 Hz, 2 H, H_{Leu}-γ), 2.00 (ddd, ^2J = 13.9, ^3J = 10.5, ^3J = 5.2 Hz, 2 H, H_{Leu}-β_b), 3.16 (dd, ^3J = 3.5, ^3J = 2.5 Hz, 2 H, H-3), 3.74 (d, ^2J = 15.8 Hz, 2 H, H_{Gly}-α_a), 3.89 (d, ^2J = 15.8 Hz, 2 H, H_{Gly}-α_b), 4.48−4.54 (m, 2 H, H_{Leu}-α), 4.51 (d, ^3J = 2.5 Hz, 2 H, H-4), 4.52 (dq, ^3J = 7.3, ^3J = 7.1 Hz, 2 H, H_{Ala}-α), 6.77 (d, ^3J = 7.1 Hz, 2 H, NH_{Ala}), 7.32−7.42 (2 × m, 10 H, Ph), 7.65 (d, ^3J = 9.2 Hz, 2 H, NH_{Leu}) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 17.79 (q, C_{Ala}-β), 21.62 (q, C_{Leu}-δ_a), 23.09 (q, C_{Leu}-δ_b), 24.94 (d, C_{Leu}-γ), 41.19 (t, C_{Leu}-β), 45.05 (d, C_{Leu}-α), 48.17 (t, C_{Gly}-α), 51.14 (d, C_{Ala}-α), 59.37 (d, C-4), 64.32 (d, C-3), 126.52, 128.79, 129.15 (3 × d, aryl-*C* Ph), 136.52 (s, aryl-*C ipso* Ph), 166.07 (s, CO_{Gly}), 169.27 (s, C-2), 172.91 (s, CO_{Ala}) ppm. $C_{38}H_{50}N_6O_6$ (686.84): calcd. C 66.45, H 7.34, N 12.24; found C 65.03, H 7.28, N 11.87. HR-MS: calcd. for $^{12}C_{38}{}^1H_{51}{}^{14}N_6{}^{16}O_6$ 687.3870; found *m/z* = 687.3873.

Tetrapeptide Analog 17a: Et_2NH (627 mg, 890 μL, 8.60 mmol) was added to a solution of the tripeptide analog **10a** (321 mg, 470 μmol) in CH_2Cl_2 (7 mL) and the mixture was stirred at room temp. for 5 h (monitored by TLC). The volatile components were removed (20 mbar, 40 °C) and the residue was repeatedly dissolved in THF and the solvents were evaporated to remove traces of Et_2NH. The residue was dissolved in THF (8 mL) and Boc−Ala−OPfp (**9**, 176 mg, 520 μmol) was added. After stirring for 2.5 h the solvent was removed (100 mbar, 40 °C), the residue was dissolved in EtOAc, extracted with brine (3×), dried ($MgSO_4$), filtered, and concentrated in vacuo (50 mbar, 40 °C). Column chromatography (SiO_2, PE/EtOAc, 2:1) yielded the tetrapeptide analog **17a** (281 mg, 450 μmol, 96%) as a colorless foam. M.p. 135−137 °C. $[\alpha]_D^{20} = -53.7$ ($c = 1.01$, $CHCl_3$). $C_{34}H_{46}N_4O_7$ (622.75): calcd. C 65.57, H 7.45, N 9.00; found C 65.12, H 7.24, N 8.85. HR-MS: calcd. for $^{12}C_{34}{}^1H_{47}{}^{14}N_4{}^{16}O_7$ 623.3445; found *m/z* = 623.3448.

Tetrapeptide Analog 17b: Et_2NH (4.29 g, 6.10 mL, 58.7 mmol) and the tripeptide analog **10b** (400 mg, 590 μmol) dissolved in acetonitrile (12 mL) were allowed to react as described for compound **17a**. Traces of Et_2NH were removed by evaporation with acetonitrile. The residue was dissolved in THF (12 mL) and reacted with Boc−Ala−OPfp (**9**, 218 mg, 650 μmol) as described above. Column chromatography (SiO_2, PE/EtOAc, 7:3) yielded the tetrapeptide analog **17b** (351 mg, 560 μmol, 95%) as a colorless foam. M.p. 92−104 °C (softening range). $[\alpha]_D^{20} = -30.9$ ($c = 1.00$, $CHCl_3$). $C_{34}H_{46}N_4O_7$ (622.75): calcd. C 65.57, H 7.45, N 9.00; found C 65.19, H 7.49, N 8.77. HR-MS: calcd. for $^{12}C_{34}{}^1H_{47}{}^{14}N_4{}^{16}O_7$ 623.3445; found *m/z* = 623.3445.

Acyclic Precursor 19a: To a solution of Boc−Ala−Ala−Phe−OMe (**18**, 227 mg, 540 μmol) in THF (30 mL), 0.2 N aqueous LiOH (90 mL) was added in 3 portions at 0 °C within 1.5 h. Stirring was continued for 20 h and the solution was added to a mixture of 0.2 N HCl (300 mL) and EtOAc (100 mL). The aqueous phase was extracted with EtOAc (2×) and the combined organic layers were dried ($MgSO_4$) and the solvent evaporated (50 mbar, 40 °C). The residue was dissolved in CH_2Cl_2 (10 mL), the solution stirred and cooled to 0 °C, then pentafluorophenol (199 mg, 1.08 mmol) and EDC (125 mg, 650 μmol) were added. Stirring was continued at 0 °C for 2 h and at room temp. for 2 h The solvents were removed (300 mbar, 35 °C), EtOAc and brine were added to the residue. The organic layer was extracted with saturated $NaHCO_3$ (2×) and with brine, dried ($MgSO_4$), filtered through a pad of Celite®, and concentrated in vacuo (50 mbar, 40 °C) to yield Boc−Ala−Ala−Phe−OPfp. During this procedure ca. 20% epimerization at C-α of Phe was observed.

To the dipeptide analog **6a** (292 mg, 540 μmol) dissolved in acetonitrile (15 mL), Et_2NH (4.74 g, 6.73 mL, 64.8 mmol) was added, and the mixture was stirred for 0.5 h at room temp. (monited by TLC). The volatile components were removed (20 mbar, 40 °C) and the residue was repeatedly dissolved in acetonitrile and the solvents were evaporated in order to remove traces of Et_2NH. The residue was diluted in CH_2Cl_2 (15 mL) and Boc−Ala−Ala−Phe−OPfp (as described above) was added. After stirring at room temp. for 2.5 h, the solvents were removed (100 mbar, 40 °C), the residue was dissolved in EtOAc, extracted with brine (3×), dried ($MgSO_4$), filtered, and concentrated in vacuo. The major isomer was partly separated by recrystallization (EtOAc), the mother liquor was then resolved by MPLC (PE/*i*PrOH, 91:9) affording the acyclic precursor **19a** (177 mg, 250 μmol, 46%), its epimer **epi-19a** (57 mg, 81 μmol, 15%) and an additional fraction containing a mixture of the isomers (35 mg, 49 μmol, 9%) as colorless solids. **Major Isomer 19a:** M.p. 214−215 °C. $[\alpha]_D^{20} = -72.0$ ($c = 0.50$, $CHCl_3$). $C_{38}H_{53}N_5O_8$ (707.86): calcd. C 64.48, H 7.55, N 9.89; found C 62.08, H 7.29, N 9.55. HR-MS: calcd. for $^{12}C_{38}{}^1H_{54}{}^{14}N_5{}^{16}O_8$ 708.3972; found *m/z* = 708.3984. **Minor Isomecr epi-19a:** M.p. 188−189 °C. $[\alpha]_D^{20} = -35.4$ ($c = 0.50$, $CHCl_3$). $C_{38}H_{53}N_5O_8$ (707.86): calcd. C 64.48, H 7.55, N 9.89; found C 64.34, H 7.47, N 9.91.

Acyclic Precursor 19b: Dipeptide analog **6b** (292 mg, 540 μmol) was deprotected and reacted with Boc−Ala−Ala−Phe−OPfp as described for compound **19a** (reaction of intermediate **6a**). Separation of side products and impurities by column chromatography (PE/EtOAc, 3:1 to 1:1) and separation of the isomers by MPLC (PE/*iPr*OH, 9:1) afforded the acyclic precursor **19b** (195 mg, 275 μmol, 51%) and its epimer **epi-19b** (65 mg, 92 μmol, 17%) as colorless solids. **Major Isomer 19b:** M.p. 180−182 °C. $[\alpha]_D^{20} = -39.5$ ($c = 1.00$, $CHCl_3$). $C_{38}H_{53}N_5O_8$ (707.86): calcd. C 64.48, H 7.55, N 9.89; found C 63.99, H 7.57, N 9.70. HR-MS: calcd. for $^{12}C_{38}{}^1H_{54}{}^{14}N_5{}^{16}O_8$ 708.3972; found *m/z* = 708.3971. **Minor Isomer epi-19b:** M.p. 186−187 °C. $[\alpha]_D^{20} = -12.2$ ($c = 0.50$, $CHCl_3$). HR-MS: calcd. for $^{12}C_{38}{}^1H_{54}{}^{14}N_5{}^{16}O_8$ 708.3972; found *m/z* = 708.3976.

Cyclic Peptidomimetic 20: Saponification of the Boc-protected acyclic precursor **17a** (205 mg, 329 μmol) in THF (25 mL) with 0.2 N aqueous LiOH (15 mL), activation with pentafluorophenol (63 mg, 0.34 mmol after addition of 15 mL CH_2Cl_2) and EDC

(79 mg, 0.41 mmol), deprotection with TFA (4.10 g, 2.75 mL, 36.0 mmol) and cyclization in vigorously stirred 1 M aqueous K_2CO_3 (18 mL)/$CHCl_3$ (36 mL) was achieved as described for compound **16**. Recrystallization afforded the cyclic peptidomimetic **20** (150 mg, 307 μmol, 93%) as a colorless solid. M.p. 225−227 °C. $[\alpha]_D^{20} = -12.7$ ($c = 1.00$, $CHCl_3$). IR (KBr): $\tilde{\nu}$ = 3310 (N−H), 3038, 3005 (arom. C−H), 2929, 2903, 2842 (aliphat. C−H), 1749, 1672, 1652 (C=O), 1520 (δ C−N−H and ν C−N, amide II), 1443 (δ-C−H and C=C), 724, 677 (arom. C−$H_{o,o,p}$) cm^{-1}. MS (FAB, pos., matrix: NBA): *m/z* = 513 (43) $[M + Na]^+$, 491 (100) $[M + H]^+$, 189 (9) $[PhCH{=}NHCH_2C(O)NC_2H_4]^+$, 131 (13) $[PhC_3H_2O]^+$, 120 (27) $[H_2N{=}CHCH_2Ph]^+$, 91 (15) $[C_7H_7]^+$. ^1H NMR (300 MHz, $CDCl_3$, 330 K): δ = 0.89 (d, ^3J = 6.5 Hz, 3 H, H_{Leu}-δ_a), 0.95 (br. d, ^3J = 6.6 Hz, 3 H, H_{Leu}-δ_b), 1.33 (d, ^3J = 6.9 Hz, 3 H, H_{Ala}-β), 1.37 (ddd, ^2J = 13.9, ^3J = 8.8, ^3J = 4.8 Hz, 1 H, H_{Leu}-β_a), 1.67 (m_c, 1 H, H_{Leu}-γ), 1.89 (ddd, ^2J = 13.9, ^3J = 10.5, ^3J = 5.4 Hz, 1 H, H_{Leu}-β_b), 3.06 (d, ^2J = 14.3 Hz, 1 H, H_{Gly}-α_a), 3.14 (dd, ^3J = 2.6, ^3J = 2.5 Hz, 1 H, H-3), 3.26 (dd, ^2J = 13.7, ^3J = 5.3 Hz, 1 H, H_{Phe}-β_a), 3.77 (m_c, 1 H, H_{Phe}-β_b), 3.96 (m_c, 1 H, H_{Phe}-α), 4.22 (d, ^2J = 14.3 Hz, 1 H, H_{Gly}-α_b), 4.35 (dq, ^3J = 9.6, ^3J = 6.9 Hz, 1 H, H_{Ala}-α), 4.42 (d, ^3J = 2.5 Hz, 1 H, H-4), 4.49 (dddd, ^3J = 10.5, ^3J = 9.7, ^3J = 4.8, ^3J = 2.6 Hz, 1 H, H_{Leu}-α), 6.57 (br. s, 1 H, NH_{Phe}), 6.70 (d, ^3J = 9.6 Hz, 1 H, NH_{Ala}), 7.18−7.39 (m, 10 H, 2 Ph) ppm, signal of NH_{Leu} is hidden. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 14.37 (q, C_{Ala}-β), 21.44, 23.22 (2 × q, C_{Leu}-δ), 25.14 (d, C_{Leu}-γ), 35.55 (t, C_{Phe}-β), 41.63 (t, C_{Leu}-β), 44.30 (d, C_{Leu}-α), 46.04 (t, C_{Gly}-α), 49.05 (d, C_{Ala}-α), 57.96 (d, C-4), 62.93 (d, C_{Phe}-α), 65.53 (d, C-3), 126.33, 126.90, 128.54, 128.78, 128.95, 129.06 (6 × d, aryl-*C* Ph), 136.51, 137.44 (2 × s, aryl-*C ipso* Ph), 169.90, 169.43, 173.24, 173.56 (4 × s, *C*=O amide, β-lactam) ppm. $C_{28}H_{34}N_4O_4$ (490.59): calcd. C 68.55, H 6.99, N 11.42; found C 67.92, H 6.96, N 11.19. HR-MS: calcd. for $^{12}C_{28}{}^1H_{35}{}^{14}N_4{}^{16}O_4$ 491.2658; found *m/z* = 491.2671.

Cyclic Peptidomimetic 21: To a solution of the Boc-protected acyclic precursor **19a** (100 mg, 141 μmol) in DMF (6.5 mL), 1 M aqueous NaOH (420 μL) was added dropwise at room temp. within 3 h. Stirring was continued for 15 h and the solution was added to a mixture of 0.5 N HCl (25 mL) and CH_2Cl_2 (45 mL). The aqueous phase was extracted with CH_2Cl_2 (3×) and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated in vacuo (200 mbar, 40 °C). Activation with pentafluorophenol (28 mg, 0.15 mmol after addition of 5 mL CH_2Cl_2) and EDC (31 mg, 0.16 mmol), deprotection with TFA (3.19 g, 2.14 mL, 28.0 mmol) and cyclization in vigorously stirred 1 M aqueous K_2CO_3 (14 mL)/$CHCl_3$ (28 mL) was achieved as described for compound **16**. Column chromatography (SiO_2, PE/EtOAc, 1:3) afforded the cyclic peptidomimetic **21** (49 mg, 85 μmol, 60%) as colorless solid. M.p. 254−256 °C. $[\alpha]_D^{20} = -94.1$ ($c = 0.80$, $CHCl_3$). IR (KBr): $\tilde{\nu}$ = 3400, 3307 (N−H), 3043, 3012 (arom. C−H), 2938, 2910, 2851 (aliphat. C−H), 1758−1740, 1672−1643 (C=O), 1521 (δ C−N−H and ν C−N, amide II), 1442 (δ C−H and C=C), 722, 677 (arom. C−$H_{o,o,p}$) cm^{-1}. MS (FAB, pos., matrix: NBA): *m/z* = 598 (18) $[M + Na]^+$, 576 (100) $[M + H]^+$, 131 (32) $[PhC_3H_2O]^+$, 120 (21) $[H_2N{=}CHCH_2Ph]^+$. Ala_I is connected to the β-lactam followed by Phe, Ala_{III}, Ala_{IV} and Leu: ^1H NMR (500 MHz, CD_2Cl_2): δ = 0.71 (d, ^3J = 6.5 Hz, 3 H, H_{Leu}-δ_a), 0.91 (d, ^3J = 6.6 Hz, 3 H, H_{Leu}-δ_b), 1.25 (d, ^3J = 6.8 Hz, 3 H, $H_{Ala\text{-}I}$-β), 1.26 (d, ^3J = 7.3 Hz, 3 H, $H_{Ala\text{-}III}$-β or $H_{Ala\text{-}IV}$-β), 1.44 (d, ^3J = 7.0 Hz, 3 H, $H_{Ala\text{-}III}$-β or $H_{Ala\text{-}IV}$-β), 1.61 (ddd, ^2J = 13.7, ^3J = 9.0, ^3J = 5.8 Hz, 1 H, H_{Leu}-β_a), 1.81 (dseptd, ^3J = 9.0, ^3J = 6.6, ^3J = 4.9 Hz, 1 H, H_{Leu}-γ), 2.38 (ddd, ^2J = 13.7, ^3J = 10.6, ^3J = 4.9 Hz, 1 H, H_{Leu}-β_b), 3.09−3.14 (m, 2 H, H_{Phe}-β), 3.14 (dd, ^3J = 3.8, ^3J = 2.6 Hz, 1 H, H-3), 3.49 (dd, ^3J = 10.6, ^3J = 5.8 Hz, 1 H, H_{Leu}-α), 3.90 (dq, ^3J = 7.2, ^3J = 7.0 Hz, 1 H, $H_{Ala\text{-}III}$-α or $H_{Ala\text{-}IV}$-α), 3.91 (dq, ^3J = 7.3, ^3J = 7.2 Hz, 1 H, $H_{Ala\text{-}III}$-α or $H_{Ala\text{-}IV}$-α), 4.10 (ddq, ^3J = 7.0, ^3J = 6.8, ^3J = 3.8 Hz, 1 H, $H_{Ala\text{-}I}$-α), 4.11 (d, ^3J = 2.6 Hz, 1 H, H-4), 4.46 (ddd, ^3J = 9.0, ^3J = 8.2, ^3J = 7.8 Hz, 1 H, H_{Phe}-α), 7.03 (d, ^3J = 7.3 Hz, 1 H, $NH_{Ala\text{-}III}$ or $NH_{Ala\text{-}IV}$), 7.17−7.23, 7.27−7.35 (2 × m, 11 H, 2 Ph, $NH_{Ala\text{-}I}$), 7.57 (d, ^3J = 9.0 Hz, 1 H, NH_{Phe}), 7.87 (d, ^3J = 7.1 Hz, $NH_{Ala\text{-}III}$ or $NH_{Ala\text{-}IV}$) ppm. ^{13}C NMR (125 MHz, CD_2Cl_2): δ = 16.32 (2 × q, $C_{Ala\text{-}III}$-β, C-IV-β), 18.62 (q, $C_{Ala\text{-}I}$-β), 21.41, 22.96 (2 × q, C_{Leu}-δ), 25.69 (d, C_{Leu}-γ), 38.20 (t, C_{Phe}-β), 40.07 (t, C_{Leu}-β), 43.91 (d, $C_{Ala\text{-}I}$-α), 50.70, 50.77 (2 × d, $C_{Ala\text{-}III}$-α, $C_{Ala\text{-}IV}$-α), 57.08 (d, C_{Phe}-α), 58.27 (d, C-4), 59.65 (d, C_{Leu}-α), 63.25 (d, C-3), 127.18, 127.31, 128.80, 129.07, 129.23, 129.62 (6 × d, aryl-*C* Ph), 137.52, 137.72 (2 × s, aryl-*C ipso* Ph), 169.65, 171.59, 171.75, 171.82, 172.91 (5 × s, 5 *C*=O) ppm. HR-MS: calcd. for $^{12}C_{32}{}^1H_{42}{}^{14}N_5{}^{16}O_5$ 576.3186; found *m/z* = 576.3201.

Polymerized Material 22: To a solution of the Boc-protected acyclic precursor **17b** (100 mg, 160 μmol) in THF (2.5 mL), 0.2 N aqueous LiOH (7.2 mL) was added in 3 portions at 0 °C within 1.5 h. Stirring was continued for 2 h and the solution was added to a mixture of 0.2 N HCl (300 mL) and EtOAc (100 mL). The aqueous phase was extracted with EtOAc (2×) and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated in vacuo (50 mbar, 40 °C). The residue was dissolved in CH_2Cl_2 (5 mL), pentafluorophenol (33 mg, 0.18 mmol) and EDC (36 mg, 0.19 mmol) were added at 0 °C. Stirring was continued for 2 h at 0 °C and for 2 h at room temp. The solvents were removed (300 mbar, 35 °C) and EtOAc and brine were added to the residue. The organic layer was extracted with saturated $NaHCO_3$ (2×) and with brine, dried ($MgSO_4$), filtered through a pad of Celite®, and the solvent was evaporated (50 mbar, 40 °C).

Deprotection was achieved by treatment of the residue with TFA (1.82 g, 1.22 mL, 16.0 mmol) at 0 °C for 2.5 h. After removal of excess TFA in vacuo (20 mbar, 40 °C) the obtained salt was dissolved in CH_2Cl_2, and cyclization was attempted by dropwise addition (1 h) of the solution to a vigorously stirred emulsion of 1 M aqueous K_2CO_3 (9 mL) and $CHCl_3$ (16 mL) as described previously. The aqueous phase was separated after further stirring for 1 h and extracted with $CHCl_3$ (3×). The combined organic layers were dried ($MgSO_4$), filtered through a pad of Celite®, and the solvent was evaporated (50 mbar, 40 °C) to leave a polymeric material **22** (95 mg) which was not further purified.

Polymerized Material 23: Saponification of the Boc-protected acyclic precursor **19b** (124 mg, 180 μmol) in THF (3 mL) with 0.2 N aqueous LiOH (7.8 mL), activation in CH_2Cl_2 (6 mL) with pentafluorophenol (33 mg, 0.18 mmol) and EDC (38 mg, 0.20 mmol), deprotection with TFA (2.09 g, 1.40 mL, 18.3 mmol) and addition to an emulsion of 1 M K_2CO_3 solution (9 mL) and $CHCl_3$ (18 mL) as described for compound **22** yielded a polymeric material **23** (128 mg) which was not further purified. MS (MALDI-TOF, pos., matrix: ATT): *m/z* = 2306 (traces, *n* = 4), 1730 (traces, *n* = 3), 1153 (35, *n* = 2), 577 (100, *n* = 1).

Supporting Information: Spectroscopic data with peak assignments for all compounds. ^1H and ^{13}C NMR spectra of compounds **15**, **16**, **17a**, **19a**, **20**, **21**. A scheme rationalizing the conformation in the vicinity of the β-lactam in compound **16**.

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