A Chirally Stable, Atropoisomeric, C^{α}-Tetrasubstituted α-Amino Acid: Incorporation into Model Peptides and Conformational Preference

by **Fernando Formaggio**, **Cristina Peggion**, **Marco Crisma**, and **Claudio Toniolo***

Biopolymer Research Centre, CNR, Department of Organic Chemistry, University of Padova, via Marzolo 1, I-35131 Padova (e-mail: biop02@chor.unipd.it)

and **Luba Tchertanov** and **Jean Guilhem**

Institut de Chimie des Substances Naturelles, UPR 2301, F-91198 Gif-sur-Yvette

and **Jean-Paul Mazaleyrat**, **Yolaine Goubard**, Anne Gaucher, and **Michel Wakselman**

SIRCOB, Bât. Lavoisier, University of Versailles, F-78035 Versailles

A variety of model peptides, including four complete homologous series, to the pentamer level, characterized by the recently proposed binaphthyl-based, axially chiral, C^{α}-tetrasubstituted, cyclic α-amino acid Bin, in combination with Ala, Gly, or Aib residues, was synthesized by solution methods and fully characterized. The solution conformational propensity of these peptides was determined by FT-IR absorption and ^{1}H-NMR techniques. Moreover, the molecular structures of the free amino acid (*S*)-enantiomer and an N^{α}-acylated dipeptide alkylamide with the heterochiral sequence -(*R*)-Bin-Phe- were assessed in the crystal state by X-ray diffraction. Taken together, the results point to the conclusion that β-bends and 3_{10} helices are preferentially adopted by Bin-containing peptides, although the fully extended conformation would also be adopted in solution by the short oligomers to some extent. We also confirmed the tendency of (*R*)-Bin to fold a peptide chain into *right*-handed bend and helical structures. The absolute configuration of the Bin residue(s) was correlated with the typically intense exciton-split *Cotton* effect of the ^1B_b binaphthyl transition near 225 nm.

Introduction. – One of the major drawbacks for the exploitation of peptides as drugs is their remarkable conformational flexibility leading to undesired interactions with different receptors. As a consequence, in recent years, conformationally constrained analogues of bioactive peptides have acquired increasing popularity among medicinal chemists, in an effort to firmly establish three-dimensional structure-bioactivity relationships and to develop new drugs with prolonged action and/or more selective properties [1–7]. In particular, conformational restriction through $C_i^{\alpha} \leftrightarrow C_i^{\alpha}$ cyclization generates the family of 1-aminocycloalkane-1-carboxylic acid (Ac_nc) residues [7].

Theoretical and experimental studies of the preferred conformations of peptides characterized by the Ac_nc ($n = 4-9$) residues have been the subject of recent review articles and papers [8–12]. In a close parallelism to the structural behaviour of Aib (α-aminoisobutyric acid) [8][13] (*Fig. 1*), the prototype of C^{α}-*tetra*substituted achiral α-amino acids and Iva (isovaline), the prototype of their chiral analogues [14–16], it was shown that regular β-bend forms [17–19] or 3_{10}-helical structures [20] are adopted as a function of main-chain length.

We now describe the synthesis and conformational propensity of peptides containing Bin (=4,5-dihydro-4-amino-3*H*-cyclohepta[2,1-*a*:3,4-*a*′]dinaphthalene-4-

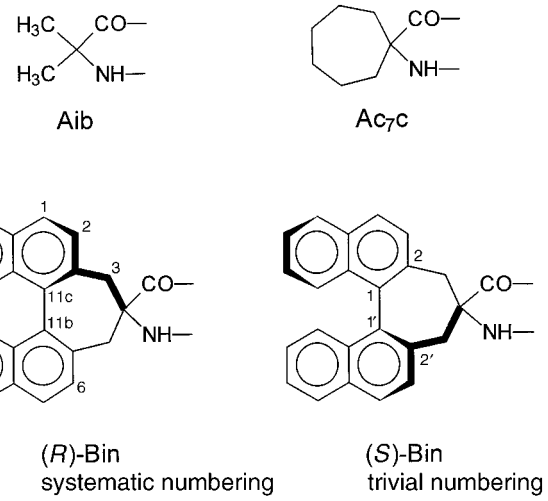

Fig. 1. *Chemical structures of Aib, Ac$_7$c and the Bin enantiomers*

carboxylic acid) (*Fig. 1*), a C$^{\alpha}$-tetrasubstituted α-amino acid related to the Ac$_n$c family, in particular to Ac$_7$c (*Fig. 1*), the seven-membered ring compound [9–11]. More specifically, four model peptide series based on the (*S*)-Bin/Ala, (*S*)-Bin/Gly, and (*S*)-Bin/Aib sequences to the pentamer level were prepared by solution methods, fully characterized, and their preferred conformation studied in solution by FT-IR absorption and ^{1}H-NMR techniques. Moreover, the crystal-structure analyses of the free amino acid itself ((*S*)-enantiomer) and a terminally blocked dipeptide with the heterochiral sequence Bz–(*R*)-Bin–Phe–NHChx (Bz, benzoyl; Chx, cyclohexyl) were also performed by X-ray diffraction. Finally, a configurational investigation by CD spectroscopy of the Bin residues was carried out.

In addition to being a member of the Ac$_n$c family, Bin is optically active, as it is characterized by an axial chirality (atropisomerism) related to the hindered rotation about the C(1)–C(1′) (=C(11b)–C(11c)) bond of the 1,1′-binapthalene moiety [21–23]. Indeed, the chain-bridged derivative Bin preserves the C_2 symmetry of 1,1′-binaphthalene (because of the presence of two identical substituents at the 2,2′-positions), but a twist is induced by the chiral binaphthalene moiety in the prochiral, fused Ac$_7$c structure [24]. Because of their highly stable configuration, 1,1′-binapthalene systems have been extensively exploited as chiral auxiliaries in stereoselective organic synthesis, in the preparation of chiral hosts for molecular-recognition studies, and in the construction of new materials [21–23][25][26]. Preliminary accounts of a limited part of this work have been published [27–30]. The results of an X-ray diffraction investigation, photophysical experiments based on the binaphthalene fluorophore, and molecular-mechanics conformational calculations of a hexapeptide containing a single, N-terminal (*S*)-Bin residue have been reported [31]. A derivative of an interesting binaphthalene C$^{\alpha}$-*tri*substituted α-amino acid, 1-(1-naphthyl)-2-naphthylalanine (=β-([1,1′-binaphthalen]-2-yl)alanine=α-amino-[1,1′-binaphthalene]-2-propanoic acid), has been prepared *via* a tandem catalysis procedure [32]. Closer Bin analogues with atropoisomeric 2,2′,6,6′-tetrasubstituted 1,1′-biphenyl

architectures, which possess either Me [33] or OH groups (precursor of crown-ether receptors) [34][35] in the 6,6′-positions, have also been described.

Results and Discussion. – *Amino-Acid and Peptide Synthesis.* The synthesis and characterization of racemic Bin, the Bin enantiomers, and their derivatives H–Bin–O^{t}Bu (O^{t}Bu, *tert*-butoxy), Boc–Bin–OH (Boc, (*tert*-butoxy)carbonyl), and Z–Bin–OH (Z, (benzyloxy)carbonyl) were already described [27–30]. Briefly, C^{α}-bis-alkylation of a *Schiff* base from H–Gly–O^{t}Bu by either racemic or enantiomerically pure 2,2′-bis(bromomethyl)-1,1′-binaphthalene under phase-transfer conditions led in good yields (*ca.* 80%) to the corresponding α-amino esters H–Bin–O^{t}Bu. Acidolysis of the amino esters with CF_3COOH/CH_2Cl_2 1:1 gave the free amino acids. The N^{α}-protected derivatives were prepared by standard methods. Direct and indirect racemate separation of the free amino acid and the N^{α}-protected derivatives were achieved by HPLC on a chiral stationary phase or by pre-column derivatization with a chiral reagent [36].

In the present work, the peptides **1**–**14** containing Bin were studied (*Table 1*). The (*S*)-Bin/Ala and (*S*)-Bin/Gly peptides were synthesized by means of the step-by-step strategy in solution starting from the C-terminus. (*S*)-Bin–Ala, (*S*)-Bin–Gly, Ala–Ala and Gly–Gly peptide-bond formation was achieved by the DCC/HOBt (DCC, *N*,*N*′-dicyclohexylcarbodiimide; HOBt, 1-hydroxy-1*H*-1,2,3-benzotriazole) method [37], while the Ala–(*S*)-Bin and Gly–(*S*)-Bin couplings were performed by the symmetrical-anhydride procedure. The N^{α}-Boc protecting group was removed on treatment with either HCl/AcOEt or CF_3COOH/CH_2Cl_2 1:1. The terminally protected (*S*)-Bin–Aib dipeptide was prepared by means of the pivaloyl (Piv) mixed-anhydride method [38][39]. The *tert*-butyl ester protecting group at the C-terminus was removed by acidolysis (CF_3COOH/CH_2Cl_2 1:1). Then, a coupling strategy from the N-terminus, involving carboxy-group activation of peptides with either Aib or (*S*)-Bin as the C-terminal residue through the oxazol-5(4*H*)-one intermediate [38][39] (prepared by intramolecular dehydration of the peptide free acid with Ac_2O at high temperature),

Table 1. *Peptides Containing Bin*

1	Ac–(*R*)-Bin–Phe–NHChx
1′	Bz–(*R*)-Bin–Phe–NHChx
2	Ac–(*S*)-Bin–Phe–NHChx
3	Boc–Gly–(*S*)-Bin–Gly–OMe
4	Boc–$(Gly)_2$–(*S*)-Bin–Gly–OMe
5	Boc–(*S*)-Bin–$(Gly)_2$–(*S*)-Bin–Gly–OMe
6	Boc–Ala–(*S*)-Bin–Ala–OMe
7	Boc–$(Ala)_2$–(*S*)-Bin–Ala–OMe
8	Boc–(*S*)-Bin–$(Ala)_2$–(*S*)-Bin–Ala–OMe
9	Z–(*S*)-Bin–$(Aib)_2$–O^{t}Bu
10	Z–(*S*)-Bin–$(Aib)_2$–(*S*)-Bin–O^{t}Bu
11	Z–(*S*)-Bin–$(Aib)_2$–(*S*)-Bin–Aib–O^{t}Bu
12	Z–$(Aib)_2$–(*S*)-Bin–O^{t}Bu
13	Z–$(Aib)_2$–(*S*)-Bin–Aib–O^{t}Bu
14	Z–$(Aib)_2$–(*S*)-Bin–$(Aib)_2$–O^{t}Bu
15	H–Phe–NHChx

was employed. Acylation of either H–Aib–O^{t}Bu or H–(*S*)-Bin–O^{t}Bu by the peptide oxazolones required several days, but, in general, this reaction gave good yields (*ca.* 80%) of the (*S*)-Bin/Aib peptides.

The synthesis and characterization of the terminally blocked dipeptides Bz–(*R*)-Bin–Phe–NHChx (**1′**), the X-ray diffraction structure of which is described below, Ac–(*R*)-Bin–Phe–NHChx (**1**), and Ac–(*S*)-Bin–Phe–NHChx (**2**) were already reported [29]. The α-amino amide H–Phe–NHChx (**15**) was used as a chiral auxiliary for the medium-scale resolution of (*RS*)-Bin.

Configurational Analysis. The electronic absorption spectrum of naphthalene consists of one main band (^1B_b) at 220 nm, accompanied by two weaker bands at 286 nm (^1L_a) and 310 nm (^1L_b). In the spectra of 1,1′-binaphthalene derivatives, the strongest UV band at *ca.* 220 nm and the corresponding, extremely intense CD exciton-split bands at 200–240 nm are associated with coupling of the two ^1B_b transitions located on different naphthalene rings [40–42]. From a study on a large variety of compounds, it was concluded that the (*S*)-enantiomer of a C_2-symmetrically substituted binaphthalene would exhibit a positive *Cotton* effect at longer wavelengths and a negative *Cotton* effect at shorter wavelengths.

However, in *open*-chain binaphthalenes, the sign of the exciton-split *Cotton* effect is also sensitive to the value of the dihedral angle θ defined by the two naphthalene planes, from (*M*)-helicity ($0 < |\theta| < 90°$, s-*cis* conformer) to (*P*)-helicity ($90° < |\theta| < 180°$, s-*trans* conformer). Therefore, the absolute configuration of open-chain binaphthalene compounds is not independently determinable from the CD data alone: in these cases, information on the dihedral angle θ is needed. Luckily, as in our Bin peptides characterized by a *bridged*-chain binaphthalene system, the θ angle is forced to be less than 90° by the fusion to the seven-membered ring, the absolute configuration only plays a role on the determination of the sign of the exciton-split *Cotton* effect at 220 nm.

Fig. 2 shows the CD spectra (200–250 nm region) of the diastereoisomeric Ac–(*R*)-Bin–Phe–NHChx (**1**) and Ac–(*S*)-Bin–Phe–NHChx (**2**) in MeOH solution. From these two CD patterns and those recorded for other Bin peptides (not shown) it is evident that the (*S*)-Bin peptides show a consistently positive ^1B_b *Cotton* effect at longer wavelengths, whereas a negative *Cotton* effect at longer wavelengths is exhibited by the (*R*)-Bin peptides. Thus, the CD technique proved to be very useful for a fast and unequivocal configurational assignment of Bin peptides. An inspection of all of the CD curves strongly support the view that neither the chiral sequence nor main-chain length (and hence, presumably, peptide conformation), nature of the aromatic *N*-blocking group (Bz, Z) and amino acid side chain (Phe) have any marked influence on their shapes and intensities. By contrast, the intensity of the exciton-split *Cotton* effect is approximately doubled in the peptides with two (*S*)-Bin residues compared to that of the peptides with only one (*S*)-Bin residue.

Solution Conformational Analysis. The preferred conformations adopted by the four terminally protected peptide series based on (*S*)-Bin in combination with Gly, Ala, or Aib residues were assessed in $CDCl_3$, a solvent of low polarity, by FT-IR absorption and ^{1}H-NMR techniques as a function of concentration (over the range 10–0.1 mM). *Figs. 3* and *4* show the FT-IR absorption spectra in the conformationally most informative N–H stretching region. In general, the spectra are characterized by main bands at 3455–3425 cm^{-1} (free, solvated N–H groups) and at 3375–3330 cm^{-1}

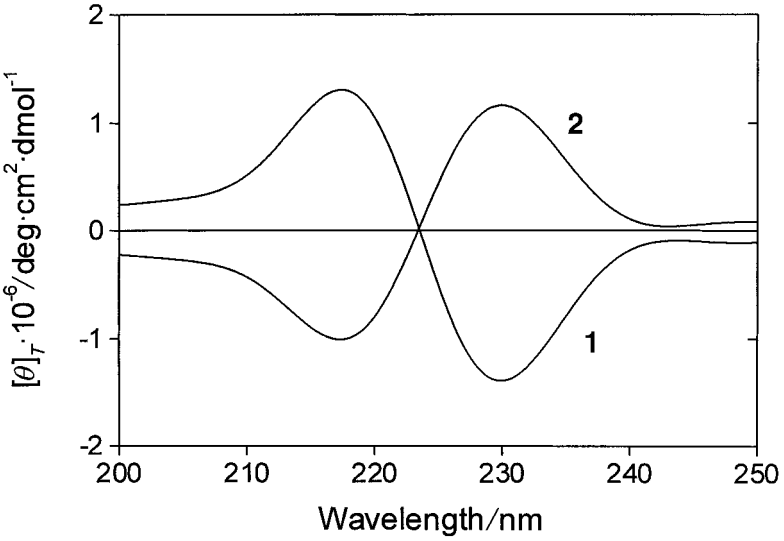

Fig. 2. *CD Spectra in the 200–250-nm region of Ac–(*R*)-Bin–Phe–NHChx* (**1**) *and Ac–(*S*)-Bin–Phe–NHChx* (**2**) *in MeOH solution.* Peptide concentration, 1.0 mM.

(strongly H-bonded N–H groups) [43–45]. The intensity of the low-frequency band relative to the high-frequency band(s) increases as the peptide main chain is elongated. This effect is quite remarkable when a (*S*)-Bin residue is incorporated at the N-terminal position of the Gly and Ala series (see **5** and **8** in *Fig. 3*), whereas the intensity enhancement is more gradual in the two Aib-rich series (*Fig. 4*). Concomitantly, the absorption maximum shifts significantly to lower wavenumbers. We were also able to demonstrate that, even at 10-mM concentration, only marginal changes take place in the spectra of the various oligomers (with the single exception of the Gly/(*S*)-Bin pentamer **5** where a variation, albeit small, is seen). Therefore, the band at 3375–3330 cm^{-1} should be interpreted as arising almost exclusively from intramolecular N–H···O=C interactions. In addition, with the exclusion of the Aib/(*S*)-Bin series **12**–**14** characterized by only one binaphthalene residue, we noted a small additional band (shoulder) in the 3415–3395 cm^{-1} region. This medium-frequency absorption is typical of weakly intramolecularly H-bonded N–H groups of fully-extended (C_5) conformers [18][46]. In a parallel analysis, the N^{α}-blocked (*S*)-Bin/Phe dipeptide alkylamides **1**, **1′**, and **2** turned out to be extensively intramolecularly H-bonded in $CDCl_3$ solution (data not shown).

The present FT-IR absorption study has provided convincing evidence that main-chain length dependent intramolecular H-bonding is a factor of paramount importance influencing the conformation of (*S*)-Bin peptides in $CDCl_3$ solution. Our results also support the view that the C^{α}-tetrasubstituted α-amino acid (*S*)-Bin, like Aib, is a much stronger inducer of intramolecularly H-bonded folded conformers than the C^{α}-di- and trisubstituted protein amino acids Gly and Ala. However, if the two (*S*)-Bin/Aib series are compared, it turns out that it is in the series **12**–**14** with only one binaphthalene residue where the regularly folded species exceedingly prevail, *i.e.*, (*S*)-Bin seems to be slightly less effective than Aib in supporting bends/helices in peptides.

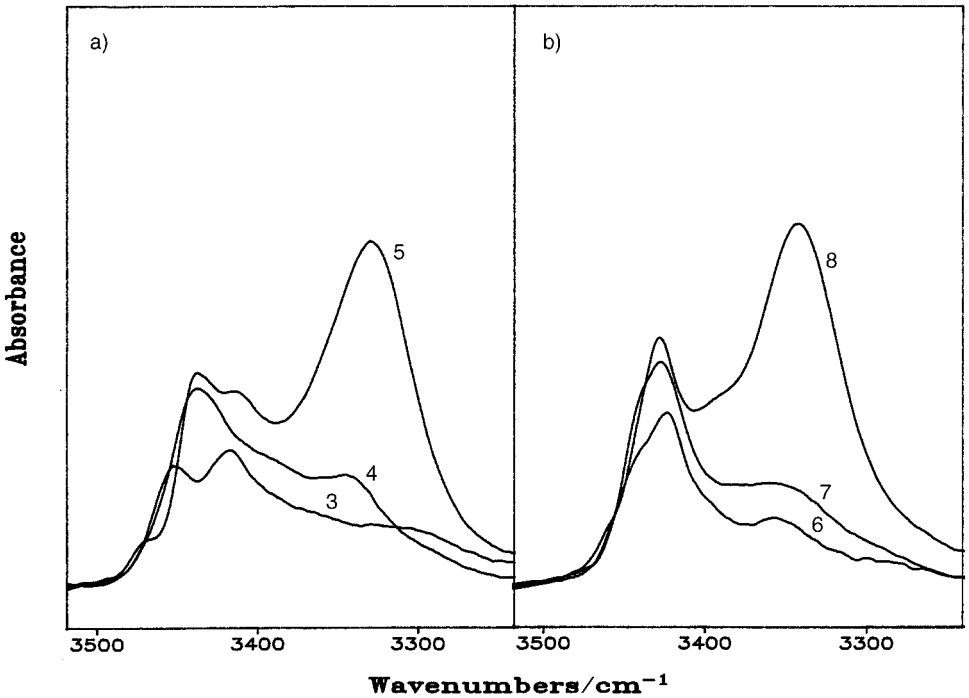

Fig. 3. *FT-IR Absorption spectra* (3500–3200-cm^{-1} region) *in* $CDCl_3$ *solution of* a) *Boc–Gly–*(S)*-Bin–Gly–OMe* (**3**), *Boc–(Gly)*$_2$*–*(S)*-Bin–Gly–OMe* (**4**), *and Boc–*(S)*-Bin–(Gly)*$_2$*–*(S)*-Bin–Gly–OMe* (**5**); b) *Boc–Ala–*(S)*-Bin–Ala–OMe* (**6**), *Boc–(Ala)*$_2$*–*(S)*-Bin–Ala–OMe* (**7**), *and Boc–*(S)*-Bin–*(Ala)$_2$*–*(S)*-Bin–*Ala–OMe (**8**). Peptide concentration, 1.0 mM.

With the aim of obtaining a more detailed information on the preferred conformations of the terminally protected (*S*)-Bin peptides in $CDCl_3$ solution, we carried out a 400-MHz ^{1}H-NMR investigation. Unfortunately, among the four pentapeptides examined, only in the case of Z–(Aib)$_2$–(*S*)-Bin–(Aib)$_2$–O^{t}Bu (**14**), all NH resonances were clearly visible, without any serious overlapping from the aromatic protons. The upfield resonance of this pentapeptide in $CDCl_3$ solution was unambiguously assigned to the urethane N(1) proton [44]. All other NH resonances were assigned by means of a 2D-ROESY experiment. From an analysis of the spectra as a function of concentration (over the 10–1.0-mM range) in $CDCl_3$ solution (results not shown), it turned out that dilution induced a small shift (0.11 ppm) to higher fields of the H–N(1)[1]) resonance. For this reason, the conformational investigation was performed at 1.0-mM peptide concentration where self-association is absent. The delineation of inaccessible (or intramolecularly H-bonded) NH groups was carried out by means of *i*) solvent dependence of NH chemical shifts on addition of increasing amounts of the strong H-bonding acceptor solvent dimethylsulfoxide (DMSO) to the $CDCl_3$ solution [47], and *ii*) free-radical TEMPO (2,2,6,6-tetramethylpiperidin-1-yloxy)-induced line broadening of NH resonances [48] (*Fig. 5*). In the pentapeptide,

[1]) The locants 1–5 refer to the position of the amino-acid residue in the peptide chain.

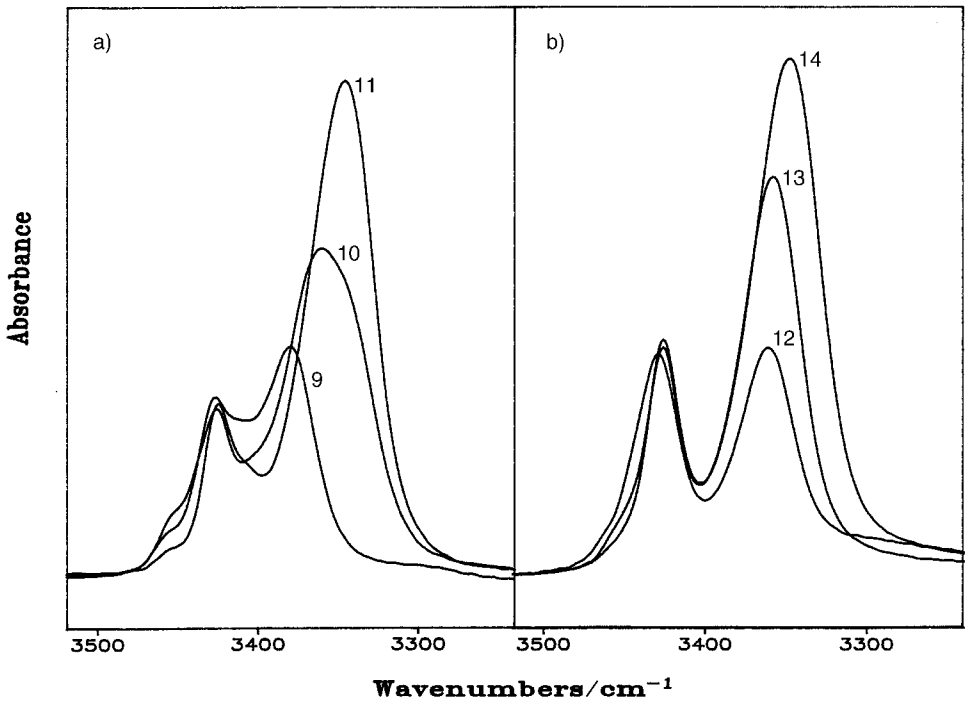

Fig. 4. *FT-IR Absorption spectra* (3500–3200-cm^{-1} region) *in* $CDCl_3$ *solution of* a) *Z*–(S)-*Bin*–$(Aib)_2$–O^t*Bu* (**9**), *Z*–(S)-*Bin*–$(Aib)_2$–(S)-*Bin*–O^t*Bu* (**10**), *and Z*–(S)-*Bin*–$(Aib)_2$–(S)-*Bin*–*Aib*-O^t*Bu* (**11**); b) *Z*–$(Aib)_2$–(S)-*Bin*–O^t*Bu* (**12**), *Z*–$(Aib)_2$–(S)-*Bin*–*Aib*–O^t*Bu* (**13**), *and Z*–$(Aib)_2$–(S)-*Bin*–$(Aib)_2$-O^t*Bu* (**14**). Peptide concentration 1.0 mM

two classes of NH protons were observed. *Class A* (H–N(1) and H–N(2)^1) includes protons whose chemical shifts are sensitive to the addition of DMSO and whose resonances broaden upon addition of TEMPO. Interestingly, the sensitivity of H–N(1) is higher than that of H–N(2). *Class B* (H–N(3) to H–N(5)^1) includes those displaying a behaviour characteristic of shielded protons (relative insensitivity of chemical shifts to solvent composition and of line widths to the presence of TEMPO).

The present ^{1}H-NMR results establish that, in $CDCl_3$ solution at 1.0 mM concentration, the H–N(3) to H–N(5)^1) protons of **14** are almost inaccessible to the perturbing agents and are, therefore, most probably, intramolecularly H-bonded. In view of the FT-IR absorption and ^{1}H-NMR findings, it is reasonable to assume that the most populated structure adopted in the structure-supporting solvent $CDCl_3$ by this terminally protected (*S*)-Bin pentapeptide **14** is the 3_{10} helix, where only the H–N(3) to H–N(5)^1) protons are involved in the intramolecular H-bonding scheme.

Two views of a molecular model of the N^{α}-acylated pentapeptide sequence –(*S*)-Bin–$(Aib)_2$–(*S*)-Bin–Aib– of **11** in the 3_{10}-helical structure are shown in *Fig. 6.* They clearly illustrate the overlapping of the two Bin side chains, one on top of the other after one complete turn of the ternary helix.

Crystal-State Conformational Analysis. The molecular and crystal structures of the free amino acid $H^+ \cdot H$–(*S*)-Bin–O^- and the terminally blocked, heterochiral

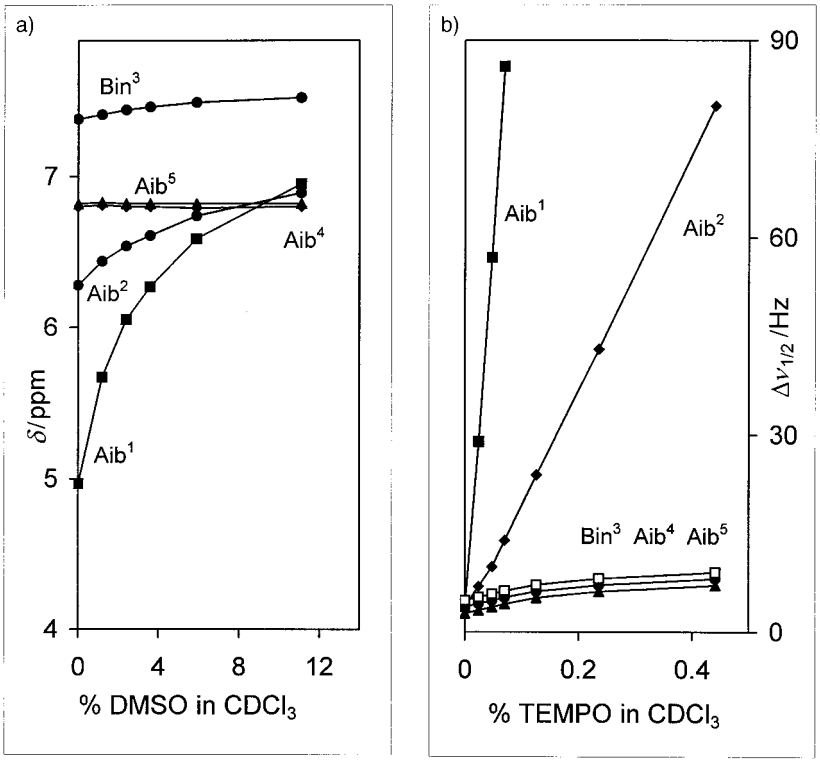

Fig. 5. a) *Plot of NH chemical shifts in the ^{1}H-NMR spectrum of Z–(Aib)$_2$–(S)-Bin–(Aib)$_2$–O^{t}Bu* (**14**) *as a function of increasing percentages of DMSO added to the CDCl$_3$ solution* (*v*/*v*). b) *Plot of bandwidths of the NH signals in the ^{1}H-NMR spectrum of* **14** *as a function of increasing percentages of TEMPO* (*w*/*v*) *added to the CDCl$_3$ solution.* Peptide concentration, 1.0 mM.

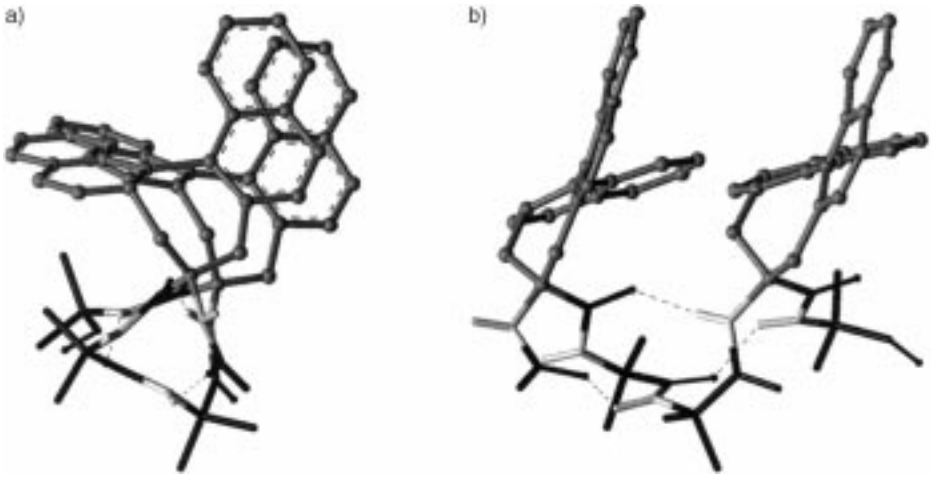

Fig. 6. *Molecular model of the 3$_{10}$-helical structure formed by the* N$^\alpha$*-acylated pentapeptide sequence –(S)-Bin–(Aib)$_2$–(S)-Bin–Aib– of* **11**. a) *Top view*; b) *side view.*

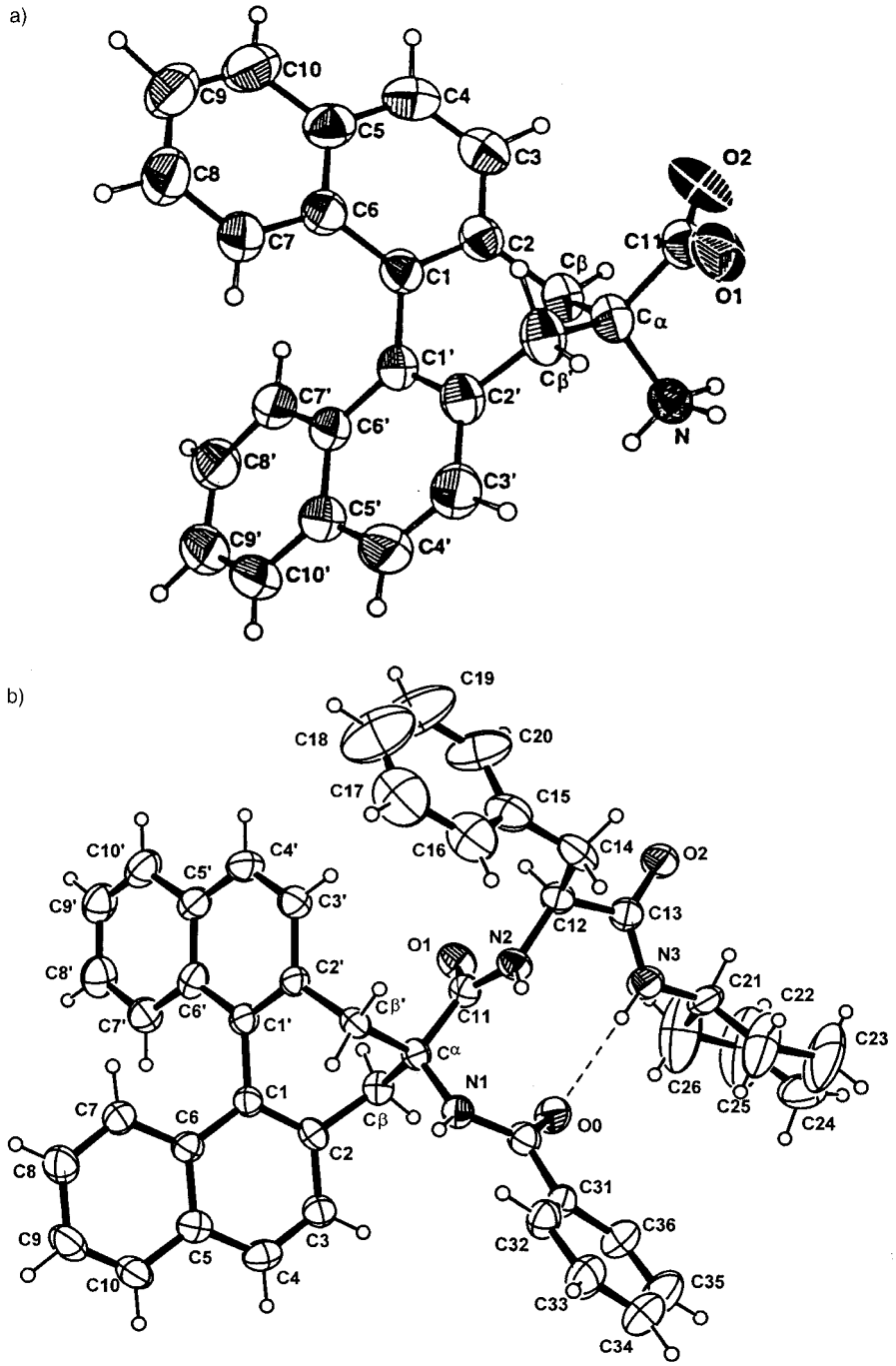

Fig. 7. *X-Ray diffraction structures of* a) $H^+ \cdot H-$(S)*-Bin−*O^- *and* b) *Bz−*(R)*-Bin−Phe−NHChx* (**1′**) *with numbering of the atoms.* The C=O···H−N intramolecular H-bond is indicated by a dashed line. Arbitrary numbering.

Table 2. *Selected Side-Chain Torsion Angles* ([°]) *of the X-Ray Diffraction Structures of H·H$_2$-Bin–O$^-$ and Its Peptides.* For numbering, see *Fig. 7*)

Torsion angle	H$^+$·H–(*S*)-Bin–O$^-$	Bz–(*R*)-Bin–Phe–NHChx (**1′**)	Boc–(*S*)-Bin–Ala–Aib–TOAC-(Ala)$_2$–O^{t}Bu[a])	
			mol. A	mol. B
C(α)–C(β)–C(2)–C(1)	–71.9(3)	74.1(6)	–74.7(5)	–72.8(5)
C(α)–C(β')–C(2′)–C(1′)	–69.4(4)	73.7(6)	–73.1(5)	–74.7(5)
C(β)–C(2)–C(1)–C(1′)	0.8(4)	2.7(8)	–1.4(6)	–1.5(6)
C(β')–C(2′)–C(1′)–C(1)	–5.4(4)	2.6(8)	–0.8(6)	–2.5(6)
C(2)–C(1)–C(1′)–C(2′)	53.8(4)	–56.5(7)	54.3(6)	55.9(6)
C(2)–C(β)–C(α)–C(β')	40.3(3)	–43.9(6)	46.3(5)	40.8(5)
C(2′)–C(β')–C(α)–C(β)	44.9(3)	–43.5(6)	40.6(5)	46.8(5)

[a]) Ref. [31].

dipeptide Bz–(*R*)-Bin–Phe–NHChx (**1′**) were determined by X-ray diffraction (see *Fig. 7*). Selected Bin side-chain torsion angles for the two structures, in comparison with those of Boc–(*S*)-Bin–Ala–Aib-TOAC–(Ala)$_2$–O^{t}Bu (TOAC, 2,2,6,6-tetramethylpiperidin-1-oxyl-4-carboxylic acid), the only published X-ray diffraction structure of a Bin-containing peptide [31], are given in *Table 2*.

The dipeptide **1′** was obtained from racemic Bz–(*RS*)-Bin–OH by reaction with optically active H–Phe–NHChx (**15**) [29], yielding 1:1 mixture of the (*R*,*S*)- and (*S*,*S*)-diastereoisomers, which were separated by column chromatography. The configuration of the binaphthalene unit of the crystalline diastereoisomer subjected to X-ray diffraction analysis was suspected to be (*R*) from the specific optical rotation by comparison with previously prepared series of similar compounds. The present X-ray diffraction data confirmed the relative configuration *rel*-(*R*,*S*) of dipeptide **1′** which is characterized by two stereogenic units, a chirality axis (Bin two-fold axis) and a chirality center (C(α)-atom of Phe).

The free amino acid is found in the dipolar (zwitterionic) form H$^+$·H–(*S*)-Bin–O$^-$ and co-crystallizes with two MeOH molecules, while the dipeptide **1′** co-crystallizes with two AcOEt molecules. The length of the characteristic C(1)–C(1′) bond joining the two naphthalene moieties of Bin (1.495(4) Å for the free amino acid and 1.501(7) Å for **1′**) compare well with those already published for the same residue [31]. In the heterochiral dipeptide **1′** all amide (peptide) moieties (ω torsion angles) are *trans*-planar [49–51]. The other backbone torsion angles [52] are: $\phi_1 = -47.6(7)^\circ$, $\psi_1 = -42.7(7)^\circ$ and $\phi_2 = -84.4(7)^\circ$, $\psi_2 = -8.4(8)^\circ$. This folded conformation, stabilized by an intramolecular H-bond C(30)=O(0)···H–N(3), the O(0)···N(3) distance (2.965(7) Å) being normal for such an interaction [53–55], is termed type-I β-bend [17–19]. The observation that the thermal ellipsoids of the atoms of this ten-membered pseudo-ring are significantly smaller than those of the remaining atoms of flexible parts of the molecule indicates rigidity of this pseudo-cyclic structure. The Phe side chain and the peptide unit are found to be in a *syn*-clinal disposition, the χ^1 torsion angle being $-72.1(7)^\circ$ [56]. The definite preference of the Phe aromatic group for $\chi^2 \approx 90^\circ$ ($105.4(9)^\circ$) is confirmed, in this conformation the ring being approximately perpendicular to the plane defined by the C(α), C(β), and C(γ) atoms. We also observed unusual C–H···π interactions in this structure. The Phe aromatic ring is in a good

position for accepting a C_{sp^2} H-atom of the Bin binaphthalene system. Actually, all H···C distances (in the range 3.570–3.336 Å) are consistent with a weak intramolecular H-bond interaction of the C(3′)–H···π type [57]. An analysis of the geometry of the Bin residues in the two structures shows that deformations may be produced in the heptacyclic system, compared to the saturated ring of the parent Ac_7c, by unfavourable steric interactions. More specifically, in each Bin residue, the seven-membered ring is pseudo-symmetrical with a non-crystallographic C_2 axis passing through the $C(\alpha)$ atom and the middle of the opposite C(1)–C(1′) bonds, the two sterically encumbering binaphthalene systems being largely non-planar (the dihedral angle between the normal to their average planes is close to 60°) [31]. The exocyclic torsion angle about the C(1)–C(1′) bond is 58.4(4)° in the (*S*)-amino acid and −62.4(8)° in the (*R*)-Bin dipeptides.

In both structures, the α-amino substituent of the Bin residue is halfway between the axial and equatorial position with respect to the average plane of the seven-membered ring.

The conformation of the seven-membered ring in the two compounds is close to a twist-boat (*TB*) [58–62], with the following puckering parameters [63]: Q_T = 1.066(3) Å, θ_2 = 84.7(2)°, ϕ_2 = 272.5(1)°, and ϕ_3 = 268.2(15)° for the free amino acid, and Q_T = 1.123(6) Å, θ_2 = 85.3(3)°, ϕ_2 = 90.0(3)°, and ϕ_3 = 88(3)° for the (*R*)-Bin dipeptide **1′**. According to literature data, in the family of conjugated cycloheptadienes, the seven-membered ring adopts a boat or a flat boat conformation [64]. The vicinal C=C bond moiety is intermediate between the *cis* and *gauche* conformations. If two aromatic rings are vicinally fused to cycloheptane, they are rotated about the joining C(1)–C(1′) bond forming a *gauche* conformation with a torsion angle in the range ±45–60°. All seven-membered rings of this type are classified as distorted boats, most of them adopting a conformation with the two torsion angles about the fusion bonds (C(β)–C(2)–C(1)–C(1′) and C(β′)–C(2′)–C(1′)–C(1)) close to 0°. Indeed, the absolute values observed for these two torsion angles in our two compounds are in the range 0.9–5.4°.

In the crystal structure of the free amino acid $H^+ \cdot H$–(*S*)-Bin–O^-, the three H-atoms of the ammonium group participate in strong H-bonds of the N–H···O type. Two of them are bound to the carboxylate O-atoms O(1) and O(2) of $(-x, y+1/2, -z)$ and $(-x, y-1/2, -z)$ symmetry-related molecules, respectively, the N(1)···O(1) and N(1)···O(2) distances being 2.701(4) and 2.856(4) Å, respectively. The third H-atom is linked to the MeOH O-atom (N···O(MeOH 2), 2.833(4) Å). Each of the two MeOH molecules doubly interacts, as donor and acceptor centres, forming O–H (MeOH 1)··· O(2)(carboxylate) (2.669(4) Å) and O–H(MeOH 2)···O(MeOH 1) (2.677(6) Å) intermolecular H-bonds [65][66]. The five different H-bonds in the crystal form a complex network of three types of ten-membered H-bonded pseudocycles.

In the β-turn-forming dipeptide **1′**, two types of intermolecular N–H···O interactions stabilize the crystal structure: N(2)–H (peptide)···O(AcOEt 1 carbonyl O-atom) (2.983(8) Å) and N(1)–H(amide)···O(2)=C(13)(amide) with the $(1-x, y+1/2, -z+1/2)$ symmetry-related dipeptide molecule, the N···O distance being 2.873(7) Å.

In summary, as far as the backbone conformation of Bin is concerned, the present crystal-state analysis confirms the published result [31] that this C^α-tetrasubstituted α-

amino acid has a marked tendency to fold, as already observed for Aib [8][13] and Iva [14–16], the prototypes of this family, and Ac_nc residues, including Ac_7c [8–12]. In addition, in analogy with the hexapeptide structure [31], also in the dipeptide discussed in this work, the relationship between amino-acid chirality and screw sense of the bend/helix formed is opposite to that commonly observed for protein amino acids, namely (*R*)-Bin forces the peptide chain to fold into *right*-handed bends/helices.

Conclusions. – The conformational tendencies of coded α-amino acids, in which chirality is exclusively associated with the presence of one (or, in few cases, two) asymmetric C-atom(s), have been extensively investigated in the last fourty years [67][68]. We now have reported for the first time a detailed analysis of the preferred conformation of a chiral α-amino acid lacking any asymmetric C-atom, the chirality of which is rather based on a different type of molecular dissymmetry, the biarene axis (pseudo-C_2 symmetry). Actually, the present study is a second step in our ongoing conformational analysis of peptides characterized by atropisomeric biarene-containing α-amino acids. In our first investigation, we have recently examined Bip, the biphenyl congener of Bin [69][70]. However, Bip undergoes racemization at room temperature which is indicative of a residual mobility in the biphenyl system. This property is responsible for the complicated conformational assignment of Bip-rich peptides, owing to the concomitant occurrence of diastereoisomeric species in their equilibrium mixtures. In contrast, Bin, a rigidified axially chiral residue is configurationally stable.

We have conclusively shown that in analogy with Aib- [8][13], Iva- [14–16], and Ac_7c-based [9–11] peptides, Bin peptides tend to fold into β-bends and 3_{10}-helices, and that the relationship between amino-acid chirality and screw sense of the bends/helices formed is opposite to that of protein amino acids. However, at least in the very short oligomers, a non-negligible population of fully extended conformers is also observed. The fact that this last property is slightly less pronounced in Bin peptides than in the corresponding Bip peptides [69][70] is possibly ascribed to the more marked chiral twist of the binaphthalene/Ac_7c fused molecular system.

Experimental Part

General. Abbreviations: DCU, *N,N'*-dicyclohexylurea; EDC, 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide; NMM; *N*-methylmorpholine. Anal. TLC and prep. column chromatography (CC): silica gel *F 254* and silica gel *60* (0.040–0.063 mm) plates (*Merck*), resp.; eluent systems: 2.5% MeOH/CH_2Cl_2 (*A*); 5% MeOH/CH_2Cl_2 (*B*); 10% MeOH/CH_2Cl_2 (*C*); UV detection (254 nm) on TLC for all compounds, even at low concentration. M.p.s: determination with a temp. raise of 3°/min; uncorrected. $[\alpha]_D$: *Perkin-Elmer 241* polarimeter, 1-dm thermostated cell. CD Spectra: *Jasco J-715* dichrograph, 1.0- and 0.2-mm fused-quartz cells; spectrograde MeOH (*Fluka*); $[\theta]_T$ (total molar ellipticity) in $^\circ \cdot cm \cdot dmol^{-1}$. FT-IR Spectra: solid-state IR by the KBr disk technique, with a *Perkin-Elmer 580-B* spectrophotometer equipped with a *Perkin-Elmer 3600* IR data station; solution IR with a *Perkin-Elmer 1720 X-FT-IR* spectrophotometer, N_2-flushed, equipped with a sample-shuttle device, at 2 cm^{-1} nominal resolution, averaging 100 scans; solvent (baseline) spectra were obtained under the same conditions; 0.1-,1.0-, and 10-mm cells with CaF_2 windows, spectrograde $CDCl_3$ (99.8% D) from *Fluka*. ^{1}H- and ^{13}C-NMR Spectra: in $CDCl_3$ (99.96% D; *Aldrich*) or (D_6)DMSO (99.96% D_6, *Acros Organics*) at 300 (400) MHz and 77 MHz, resp. *Bruker AC-300* or *-AM-400* spectrometer; δ in ppm rel. to $SiMe_4$ as internal standard, *J* in Hz; TEMPO from *Sigma.* MS: *m/z* (rel. T).

Boc–(S)-Bin–Ala–OMe. To a suspension of Boc–(*S*)-Bin–OH [28] (0.113 g, 0.25 mmol), HCl·H–Ala–OMe (0.070 g, 0.5 mmol)), and HOBt (0.068 g, 0.5 mmol) in THF (2 ml) and CH_2Cl_2 (2 ml), a soln. of Et_3N (0.051 g, 0.5 mmol) in CH_2Cl_2 (0.5 ml) and then a soln. of DCC (0.064 g, 0.3 mmol) in CH_2Cl_2 (0.5 ml) were added. The mixture was magnetically stirred at r.t. overnight and then evaporated. The residue was stirred

for a few min in the presence of AcOEt (50 ml) and the insoluble solid (DCU) filtered off. The soln. was extracted with 0.5N HCl (2 × 100 ml), H_2O (100 ml), 5% $NaHCO_3$ soln. (2 × 100 ml), and H_2O (2 × 100 ml), dried ($MgSO_4$) and evaporated and the crude product submitted to CC (column 1.5 × 41 cm, silica gel, *B*). The chromatographically pure sample was crystallized from CH_2Cl_2/hexane: 0.129 g (96%) of pure solid dipeptide. M.p. 141°. $[\alpha]_{589}^{25} = -49$, $[\alpha]_{578}^{25} = -52$, $[\alpha]_{546}^{25} = -65$, $[\alpha]_{436}^{25} = -199$, $[\alpha]_{365}^{25} = -947$ ($c = 1$, MeOH). R_f 0.75 (*B*). ^{1}H-NMR: 7.96–7.19 (*m*, 12 arom. H); 7.11 (br. *m*, NH (Ala)); 4.70 (*s*, NH (Bin)); 4.63 (*dq*, $J = 7.3$, 7.3, H–C(α)(Ala)); 3.79 (*s*, MeO); 3.41–2.33 (*dd*, $J = 12.9$, H–C(β) (Bin)); 3.31–3.12 (br. *dd*, $J \approx 13.4$, H–C(β) (Bin)); 1.49 (*s*, Boc); 1.40 (*d*, $J = 7.2$, H–C(β) (Ala)). Anal. calc. for $C_{33}H_{34}N_2O_5 \cdot 0.5\ H_2O$ (547.626): C 72.37, H 6.44, N 5.11; found: C 72.12, H 6.38, N 4.91.

H–(S)-Bin–Ala–OMe. To Boc–(*S*)-Bin–Ala–OMe (0.118 g, 0.22 mmol) in AcOEt (4 ml), 4.8N HCl in AcOEt (4 ml) was added. The resulting soln. was stirred at r.t. for 2 h and then evaporated. The residue was dissolved in AcOEt and the soln. extracted as described above for the Boc-dipeptide. The crude product was submitted to CC (column 1.5 × 41 cm, silica gel, *C*): 0.083 g (86%) of pure dipeptide. Solid foam. $[\alpha]_{589}^{25} = +165$, $[\alpha]_{578}^{25} = +171$, $[\alpha]_{546}^{25} = +192$, $[\alpha]_{436}^{25} = +264$, $[\alpha]_{365}^{25} = -235$ ($c = 0.5$, MeOH). R_f 0.45 (*B*), 0.75 (*C*). ^{1}H-NMR: 7.97–7.91 (*m*, 4 arom. H); 7.82 (*d*, $J = 7.7$, NH (Ala)); 7.58–7.19 (*m*, 8 arom. H); 4.59 (*dq*, $J = 7.3$, 7.3, H–C(α) (Ala)); 3.81 (*s*, MeO); 3.39–2.33 (*dd*, $J = 13.1$, 2 H–C(β) (Bin)); 3.11–2.48 (*dd*, $J = 13.2$, 2 H–C(β) (Bin)); 2.08 (*s*, NH_2(Bin)); 1.45 (*d*, $J = 7.2$, Me(β) (Ala)). ^{13}C-NMR: 175.0, 173.5 (C=O (Ala, Bin)); 136.3–124.8 (arom. C); 68.6 (C(α) (Bin)); 52.3 (MeO); 47.7 (Cα) (Ala)); 45.3 (C(β) (Bin)); 43.3 (Cβ') (Bin)); 17.9 (C(β) (Ala)). Anal. calc. for $C_{28}H_{26}N_2O_3 \cdot 0.3\ H_2O$ (452.917): C 74.25, H 6.14, N 6.18; found: C 74.32, H 6.06, N 5.99.

Z–Ala–(S)-Bin–Ala–OMe. To an ice-cold soln. of Z–Ala–OH (0.165 g, 0.74 mmol) in MeCN (2 ml), a soln. of DCC (0.076 g, 0.37 mmol) in MeCN (0.5 ml) was aded. The mixture was stirred at 0° for 1 h, filtered through glass wool for elimination of the DCU precipitate, and added to an ice-cold soln. of H–(*S*)-Bin–Ala–OMe (0.081 g, 0.18 mmol) in MeCN (2 ml). The resulting soln. was stirred from 0° to r.t. overnight and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. The crude product was submitted to CC (column 1.5 × 41 cm, silica gel, *C*). The resulting chromatographically pure sample was crystallized from CH_2Cl_2/hexane: 0.100 g (84%) of tripeptide. Amorphous solid. M.p. 216°. $[\alpha]_{589}^{25} = -68$, $[\alpha]_{578}^{25} = -72$, $[\alpha]_{546}^{25} = -86$, $[\alpha]_{436}^{25} = -222$, $[\alpha]_{365}^{25} = -862$ ($c = 0.5$, MeOH). R_f 0.70 (*C*). ^{1}H-NMR (Z–Ala^1-Bin–Ala^2–OMe): 7.92–7.86 (*m*, 4 arom. H); 7.55–7.22 (*m*, 13 arom. H); 7.20 (*d*, $J \approx 7.2$, NH (Ala^2), partly masked, identified by 2D COSY); 6.10 (*s*, NH (Bin)); 5.28 (*d*, $J = 6.5$, NH (Ala^1)); 5.09–5.01 (*dd*, $J = 12.2$, CH_2 (Z)); 4.53 (*dq*, $J = 7.2$, 7.2, H–C(α) Ala^2)); 4.01 (br. *dq*, $J \approx 6.8$, 6.8, H–C(α) (Ala^1)); 3.76 (*s*, MeO); 3.37–2.39 (*dd*, $J \approx 12.8$, 2 H–C(β) (Bin)); 3.43–3.15 (*dd*, $J \approx 13.5$, 2 H–C(β) (Bin)); 1.42 (*d*, $J = 7.0$, Me(β) (Ala^1)); 1.36 (*d*, $J = 7.2$, Me(β) (Ala^2)). ^{13}C-NMR: 173.7, 172.2, 171.0 (C=O (Ala^1, Ala^2, Bin)); 156.2 (C=O (Z)); 135.9–125.2 (arom. C); 70.7 (C(α) (Bin)); 67.2 (CH_2 (Z)); 52.3 (MeO); 51.4, 48.3 (C(α) (Ala^1, Ala^2)); 42.6 (C(β) (Bin)); 36.1 (C(β') (Bin)); 17.5 (C(β) (Ala^1, Ala^2)). Anal. calc. for $C_{39}H_{37}N_3O_6 \cdot 0.5\ H_2O$ (652.718): C 71.76, H 5.87, N 6.43; found: C 71.76, H 5.88, N 6.21.

Z–(S)-Bin–Ala–Ala–OMe. To a suspension of Z–(*S*)-Bin–OH [28] (0.122 g, 0.25 mmol), HCl · H–Ala–Ala–OMe (0.105 g, 0.5 mmol) and HOBt (0.068 g, 0.5 mmol) in THF (2 ml) and CH_2Cl_2 (2 ml), a soln. of Et_3N (0.051 g, 0.5 mmol) in CH_2Cl_2 (1 ml) was added, then a soln. of DCC (0.065 g, 0.3 mmol) in CH_2Cl_2 (1 ml). The mixture was magnetically stirred at r.t. overnight and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. Repeated CC (silica gel, *C*) of the crude product, followed by crystallization from CH_2Cl_2/hexane gave 0.130 g (81%) of pure tripeptide. Solid. M.p. 152°. $[\alpha]_{589}^{25} = -66$, $[\alpha]_{578}^{25} = -70$, $[\alpha]_{546}^{25} = -84$, $[\alpha]_{436}^{25} = -222$, $[\alpha]_{365}^{25} = -880$ ($c = 0.6$, MeOH). R_f 0.70 (*C*). ^{1}H-NMR (Z–Bin–Ala^1–Ala^2–OMe): 7.94–7.85 (*m*, 4 arom. H); 7.55–7.22 (*m*, 13 arom. H); 7.07 (br. *m*, NH (Ala^1)); 6.66 (*d*, $J = 7.4$, NH (Ala^2)); 5.19–5.04 (*dd*, $J = 12.1$, CH_2 (Z)); 5.08 (*s*, NH (Bin)); 4.54 (*dq*, $J = 7.2$, 7.2, H–C(α) (Ala^2)); 4.51 (br. *m*, H–C(α) (Ala^1)); 3.75 (*s*, MeO); 3.45–2.33 (*dd*, $J \approx 12.8$, 2 H–C(β) (Bin)); 3.14–3.07 (*dd*, $J \approx 13.3$, 2 H–C(β) (Bin)); 1.42 (*d*, $J = 7.2$, Me(β) (Ala^2)); 1.34 (*d*, $J = 7.0$, (Ala^1) Me(β)). ^{13}C-NMR: 173.1, 171.8 (C=O (Ala^1, Ala^2, Bin)); 155.4 (C=O (Z)); 135.9–125.3 (arom. C); 70.3 (C(α) (Bin)); 67.2 (CH_2 (Z)); 52.4 (MeO); 49.1, 48.2 (C(α) (Ala^1, Ala^2)); 42.3 (C(β) (Bin)); 37.9 (C(β') (Bin)); 17.8, 17.6 (C(β) (Ala^1, Ala^2)). Anal. calc. for $C_{39}H_{37}N_3O_6 \cdot 0.5\ H_2O$ (652.718): C 71.76, H 5.87, N 6.43; C 71.79, H 5.94, N 6.22.

Boc–Ala–(S)-Bin–Ala–OMe (**6**). To a soln. of Boc–(*S*)-Bin–Ala–OMe (0.168 g, 0.31 mmol) in CH_2Cl_2 (5 ml), CF_3COOH (5 ml) was added. The soln. was stirred at r.t. for 3 h and then evaporated. The residue was solubilized in AcOEt (100 ml). The soln. was extracted with 5% $NaHCO_3$ soln. (100 ml), dried ($MgSO_4$), and evaporated. The crude H–(*S*)-Bin–Ala–OMe (0.137 g, 0.31 mmol) was dissolved in MeCN (4 ml) and the soln. cooled to −5° and added to a cold soln. of (Boc–Ala)$_2$O, previously prepared by stirring a soln. of Boc–Ala–OH (0.236 g, 1.25 mmol) and EDC (0.120 g, 0.63 mmol) in MeCN (6 ml) at −5° for 1 h. The

mixture was magnetically stirred from $-5°$ to r.t. for 24 h and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (cilica gel, *B*) of the crude product gave 0.154 g (81%) of pure **6**. Solid. M.p. 142°. $[\alpha]_{589}^{25} = -62$, $[\alpha]_{578}^{25} = -65$, $[\alpha]_{546}^{25} = -81$, $[\alpha]_{436}^{25} = -218$, $[\alpha]_{365}^{25} = -887$ ($c = 0.2$, MeOH). R_f 0.50 (*B*). ^{1}H-NMR (Boc–Ala^1–Bin–Ala^2–OMe): 7.93–7.87 (*m*, 4 arom. H); 7.56–7.18 (*m*, 9 arom. H, masked NH (Ala^2)); 6.29 (*s*, NH (Bin)); 5.04 (*d*, $J = 6.4$, NH (Ala^1)); 4.55 (*dq*, $J = 7.2, 7.2$, H–C(α) (Ala^2)); 3.97 (*dq*, $J = 6.8, 6.8$, H–C(α) (Ala^1)); 3.75 (*s*, MeO); 3.44–3.13 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 3.38–2.41 (*dd*, $J \approx 12.7$, 2 H–C(β) (Bin)); 1.40 (*d*, $J = 7.2$, Me(β) (Ala)); 1.39 (*d*, $J = 7.3$, Me(β) (Ala)); 1.36 (*s*, Boc). ^{13}C-NMR: 173.6, 172.5, 171.2 (C=O (Ala^1, Ala^2, Bin)); 155.7 (C=O (Boc)); 134.7–125.2 (arom. C); 80.5 (Boc); 70.6 (C(α) (Bin)); 52.2 (MeO); 50.9, 48.3 (C(α) (Ala^1, Ala^2)); 42.5 (C(β) (Bin)); 36.3 (C(β') (Bin)); 28.2 (Boc); 17.7, 17.6 (C(β) (Ala^1, Ala^2)). Anal. calc. for $C_{36}H_{39}N_3O_6 \cdot 0.5\ H_2O$ (618.704): C 69.88, H 6.52, N 6.79; found: C 69.88, H 6.57, N 7.01.

*Boc–Ala–Ala–(*S*)-Bin–Ala–OMe* (**7**). Boc–Ala-(*S*)-Bin–Ala–OMe (**6**; 0.142 g, 0.23 mmol) was *N*-deprotected in CH_2Cl_2 (5 ml) and CF_3COOH (5 ml) as described above for Boc–Ala–(*S*)-Bin–Ala–OMe. A soln. of a mixture of the crude H–Ala–(*S*)-Bin–Ala–OMe (0.119 g, 0.23 mmol), Boc–Ala–OH (0.066 g, 0.35 mmol), and HOBt (0.062 g, 0.47 mmol) in THF (5 ml) and CH_2Cl_2 (2.5 ml) was cooled to $-5°$, and a soln. of EDC (0.067 g, 0.35 mmol) in CH_2Cl_2 (2.5 ml) was added. The mixture was magnetically stirred from $-5°$ to r.t. for 24 h and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (silica gel, *C*) of the crude product gave 0.148 g (93%) of pure **7**. Solid. M.p. 131°. $[\alpha]_{589}^{25} = -109$, $[\alpha]_{578}^{25} = -118$, $[\alpha]_{546}^{25} = -139$, $[\alpha]_{436}^{25} = -336$, $[\alpha]_{365}^{25} = -1449$ ($c = 0.2$, MeOH). R_f 0.50 (*C*). ^{1}H-NMR (Boc–Ala^1–Ala^2–Bin–Ala^3–OMe): 7.91–7.85 (*m*, 4 arom. H); 7.52–7.16 (*m*, 9 arom. H, masked NH (Ala^3)); 6.99 (br. *d*, NH (Ala^2)); 6.64 (*s*, NH (Bin)); 5.08 (*d*, $J = 7.2$, NH (Ala^1)); 4.50 (*dq*, $J = 7.2, 7.2$, H–C(α) (Ala^3)); 4.17 (*dq*, $J = 6.6$, H–C(α) (Ala^2)); 4.08 (br. *m*, H–C(α) (Ala^1)); 3.71 (*s*, MeO); 3.40–3.09 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 3.29–2.47 (*dd*, $J \approx 12.8$, 2 H–C(β) (Bin)); 1.42 (*d*, $J = 7.0$, Me(β) (Ala)); 1.35 (*d*, $J = 7.3$, Me(β) (Ala)); 1.22 (*d*, $J = 7.0$, Me(β) (Ala)); 1.32 (*s*, Boc). ^{13}C-NMR: 173.8, 173.2, 172.0, 171.2 (C=O (Ala^1, Ala^2, Ala^3, Bin)); 155.4 (C=O Boc)); 135.0–125.2 (arom. C); 80.3 (Boc); 70.6 (C(α) (Bin)); 52.3 (MeO); 52.2, 50.9, 48.3 (C(α) (Ala^1, Ala^2, Ala^3)); 42.4 (C(β) (Bin)); 35.7 (C(β') (Bin)); 28.2 (Boc), 17.8, 17.5, 17.4 (C(β) (Ala^1, Ala^2, Ala^3)). ESI-MS (pos.): 681 (17, $[M+H]^+$), 703 (100, $[M+Na]^+$), 719 (8, $[M+K]^+$). Anal. calc. for $C_{39}H_{44}N_4O_7 \cdot H_2O$ (698.790): C 67.03, H 6.63, N 8.02; found: C 66.94, H 6.43, N 7.82.

*Boc–(*S*)-Bin–Ala–Ala–(*S*)-Bin–Ala–OMe* (**8**). Boc–Ala–Ala–(*S*)-Bin–Ala–OMe (**7**; 0.135 g, 0.20 mmol) was *N*-deprotected in CH_2Cl_2 (5 ml) and CF_3COOH (5 ml) as described above for the synthesis of Boc–Ala–(*S*)-Bin–Ala–OMe. To a soln. of crude H–Ala–Ala–(*S*)-Bin–Ala–OMe (0.110 g, 0.19 mmol), Boc–(*S*)-Bin–OH (0.086 g, 0.19 mmol), and HOBt (0.038 g, 0.28 mmol) in THF (10 ml) and CH_2Cl_2 (5 ml), a soln. of EDC (0.043 g, 0.23 mmol) in CH_2Cl_2 (5 ml) was added. The mixture was magnetically stirred at r.t. for 48 h and evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (silica gel, *B*) of the crude product, followed by crystallization from MeCN, gave 0.098 g (49%) of pure **8**. Crystals. M.p. 220°. $[\alpha]_{589}^{25} = -89$, $[\alpha]_{578}^{25} = -94$, $[\alpha]_{546}^{25} = -115$, $[\alpha]_{436}^{25} = -297$, $[\alpha]_{365}^{25} = -1139$ ($c = 0.2$, MeOH). R_f 0.55 (*C*). ^{1}H-NMR (Boc–Bin^1–Ala^2–Ala^3–Bin^4–Ala^5–OMe): 8.00–7.13 (*m*, 24 arom. H); 7.45 (masked *d*, identified by 2D COSY, NH (Ala^2, NOE with NH (Bin^1)); 7.32 (masked *d*, identified by 2D COSY, NH (Ala^5)); 6.67 (*s*, NH (Bin^4)); 6.51 (*d*, $J = 6.5$, NH (Ala^3)); 4.95 (*s*, NH (Bin^1)); 4.57 (*dq*, $J \approx 7.2, 7.2$, H–C(α) (Ala^5)); 4.14 (*dq*, $J \approx 6.6, 6.6$, H–C(α) (Ala^2), H–C(α) (Ala^3)); 3.71 (*s*, MeO); 3.57–2.29 (*dd*, $J \approx 12.7$, 2 H–C(β) (Bin)); 3.54–2.65 (*dd*, $J \approx 13.0$, 2 H–C(β) (Bin)); 3.52–2.23 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 2.97–2.90 (*dd*, $J \approx 13.0$, 2 H–C(β) (Bin)); 1.50 (*d*, $J = 7.2$, Me(β) (Ala)); 1.41 (*d*, $J = 7.3$, Me(β) (Ala^5)); 1.33 (*d*, $J = 7.3$, Me(β)); 1.40 (*s*, Boc). ^{13}C-NMR: 173.9, 172.9, 172.5, 172.1, 171.7 (C=O (Ala^2, Ala^3, Ala^5, Bin^1, Bin^4)); 155.6 (C=O (Boc)); 135.2–125.0 (arom. C); 81.4 (Boc); 70.9, 70.0 (C(α) (Bin^1, Bin^4)); 52.2 (MeO); 51.0, 49.8, 48.2 (C(α) (Ala^2, Ala^3, Ala^5)); 42.6, 41.5, 39.4, 36.4 (C(β), C(β') (Bin^1, Bin^4)); 28.2 (Boc); 17.5, 16.9, 16.5 (C(β) (Ala^2, Ala^3, Ala^5). ESI-MS (pos.): 1038 (100, $[M+Na]^+$), 1016 (29, $[M+H]^+$). Anal. calc. for $C_{63}H_{61}N_5O_8 \cdot H_2O$ (1034.174): C 73.16, H 6.14, N 6.77; found: C 73.23, H 6.14, N 6.72.

*Boc–(*S*)-Bin–Gly–OMe.* To a suspension of Boc–(*S*)-Bin–OH (0.181 g, 0.40 mmol), HCl·H–Gly–OMe (0.100 g, 0.80 mmol) and HOBt (0.108 g, 0.80 mmol) in THF (5 ml) and CH_2Cl_2 (5 ml), a soln. of CF_3COOH (0.081 g, 0.80 mmol) in CH_2Cl_2 (5 ml) was added and then a soln. of EDC (0.095 g, 0.50 mmol) in CH_2Cl_2 (5 ml). The mixture was magnetically stirred at r.t. overnight and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (silica gel, *A*) of the crude product gave 0.203 g (97%) of pure dipeptide. Solid. M.p. 164°. $[\alpha]_{589}^{25} = +127$, $[\alpha]_{578}^{25} = +128$, $[\alpha]_{546}^{25} = +143$, $[\alpha]_{436}^{25} = +171$, $[\alpha]_{365}^{25} = +371$ ($c = 0.3$, $CHCl_3$). R_f 0.25 (*B*). ^{1}H-NMR: 7.95 (*d*, $J = 8.3$, 2 arom. H); 7.90 (*d*, $J = 8.4$, 2 arom. H); 7.63 (*d*, $J = 8.3$, 1 arom. H); 7.48–7.18 (*m*, 8 H, arom. H, masked NH

(Gly)); 4.78 (br. *s*, NH (Bin)); 4.19–3.99 (2*dd*, $J = 18.2, 5.7–18.2, 5.0$, 2 H–C(α) (Gly)); 3.75 (*s*, MeO); 3.47–2.34 (*dd*, $J \approx 12.8$, 2 H–C(β) (Bin)); 3.34–3.10 (br. *dd*, $J \approx 13.1$, 2 H–C(β) (Bin)); 1.48 (*s*, Boc). ^{13}C-NMR: 172.9, 170.5 (C=O (Gly, Bin)); 154.8 (C=O (Boc)); 134.7–125.1 (arom. C); 82.3 (Boc); 70.5 (C(α) (Bin)); 52.2 (MeO); 42.4 (C(α) (Gly)); 41.4, 37.5 (C(β), C(β') (Bin)); 28.3 (Boc). Anal. calc. for $C_{32}H_{32}N_2O_5$ (524.592): C 73.26, H 6.15, N 5.34; found: C 73.22, H 6.28, N 5.24.

Boc–Gly–(S)-Bin–Gly–OMe (**3**). To a soln. of Boc–(*S*)-Bin–Gly–OMe (0.196 g, 0.37 mmol) in CH_2Cl_2 (5 ml), CF_3COOH (5 ml) was added. The soln. was stirred at r.t. for 3 h and evaporated. The residue was solubilized in AcOEt (100 ml). The soln. was extracted wtih 5% $NaHCO_3$ soln. (100 ml), dried ($MgSO_4$), and evaporated. The crude H–(*S*)-Bin–Gly–OMe (0.157 g, 0.37 mmol) was dissolved in MeCN (5 ml) and CH_2Cl_2 (5 ml). This soln. was added to a soln. of $(Boc–Gly)_2O$ previously prepared by stirring a soln. of Boc–Gly–OH (0.262 g, 1.50 mmol) and EDC (0.143 g, 0.74 mmol) in MeCN (10 ml) for 1 h. The mixture was magnetically stirred at r.t. for 24 h and then evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. The crude product was purified by crystallization from CH_2Cl_2/cyclohexane to give 0.184 g (85%) of pure **3**. Crystals. M.p. 150°. $[\alpha]_{589}^{25} = -43$, $[\alpha]_{578}^{25} = -46$, $[\alpha]_{546}^{25} = -58$, $[\alpha]_{436}^{25} = -174$, $[\alpha]_{365}^{25} = -808$ ($c = 0.2$, MeOH). R_f 0.25 (*B*). ^{1}H-NMR (Boc–Gly^1–Bin–Gly^2–OMe): 7.95–7.89 (*m*, 4 arom. H); 7.63 (*d*, $J = 8.4$, 1 arom. H); 7.48–7.19 (*m*, 8 arom. H, masked NH (Gly^2)); 6.38 (*s*, NH (Bin)); 5.11 (br. *t*, NH (Gly^1)); 4.17–3.94 (2*dd*, $J = 18.2, 5.5–18.2, 5.2$, 2 H–C(α) (Gly^2)); 3.78–3.68 (2*dd*, $J = 16.3, 5.7–16.3, 6.1$, 2 H–C(α) (Gly^1)); 3.76 (*s*, MeO); 3.54–2.40 (br. *dd*, $J \approx 12.8$, 2 H–C(β) (Bin)); 3.41–3.13 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 1.37 (*s*, Boc). ^{13}C-NMR: 172.2, 170.5, 169.9 (C=O (Gly^1, Gly^2, Bin)); 155.0 (C=O (Boc)); 134.7–125.3 (arom. C); 81.0 (Boc); 70.9 (C(α) (Bin)); 52.2 (MeO); 45.4, 42.3 (C(α) (Gly^1, Gly^2)); 41.3, 37.3 (C(β), C(β') (Bin)); 28.1 (Boc). Anal. calc. for $C_{34}H_{35}N_3O_6 \cdot 0.5\ H_2O$ (590.652): C 69.13, H 6.14, N 7.11; found: C 69.14, H 6.35, N 7.05.

Boc–Gly–Gly–(S)-Bin–Gly–OMe (**4**). Boc–Gly–(*S*)-Bin–Gly–OMe (**3**; 0.167 g, 0.29 mmol) was *N*-deprotected in CH_2Cl_2 (5 ml) and CF_3COOH (5 ml) as described above for the synthesis of Boc–Gly–(*S*)-Bin–Gly–OMe. To a soln. of crude H–Gly–(*S*)-Bin–Gly–OMe (0.138 g, 0.29 mmol), Boc–Gly–OH (0.075 g, 0.43 mmol), and HOBt (0.062 g, 0.47 mmol) in THF (5 ml) and CH_2Cl_2 (2.5 ml), a soln. of EDC (0.082 g, 0.43 mmol) in CH_2Cl_2 (2.5 ml) was added. The mixture was magnetically stirred at r.t. for 24 h and evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (silica gel, *B*) of the crude product gave 0.080 g (44%) of pure **4**. Solid. M.p. 183°. $[\alpha]_{589}^{25} = -39$, $[\alpha]_{578}^{25} = -44$, $[\alpha]_{546}^{25} = -53$, $[\alpha]_{436}^{25} = -163$, $[\alpha]_{365}^{25} = -757$ ($c = 0.2$, MeOH). R_f 0.35 (*C*). ^{1}H-NMR (Boc–Gly^1–Gly^2–Bin–Gly^3–OMe): 7.90–7.81 (*m*, 4 arom. H); 7.57 (*d*, $J = 8.3$, 1 arom. H); 7.50–7.17 (*m*, 10 H, arom. H, masked NH (Gly^2), NH (Gly^3), NH (Bin)); 5.42 (br. *t*, NH (Gly^1)); 4.10–3.84 (*dd*, partly masked *dd*, $J = 17.6, 6.1–17.6, 5.1$, 2 H–C(α) (Gly^3)); 3.84–3.73 (2 partly masked *dd*, 2 H–C(α) (Gly^2)); 3.64–3.51 (2 masked *dd*, $J \approx 16.8, 5.9$, 2 H–C(α) (Gly^1)); 3.65 (*s*, MeO); 3.37–2.62 (*dd*, $J \approx 13.1$, 2 H–C(β) (Bin)); 3.32–3.16 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 1.08 (*s*, Boc). ^{13}C-NMR: 173.0, 171.4, 170.6, 169.2 (C=O (Gly^1, Gly^2, Gly^3, Bin)); 156.4 (C=O (Boc)); 134.8–125.2 (arom. C); 80.5 (Boc); 70.8 (C(α) (Bin)); 52.1 (MeO); 44.4, 43.9, 42.1 (C(α) (Gly^1, Gly^2, Gly^3)); 41.3, 36.7 (C(β), C(β') (Bin)); 27.9 (Boc). ESI-MS (pos.): 677 (9, $[M+K]^+$), 661 (100, $[M+Na]^+$), 639 (13, $[M+H]$). Anal. calc. for $C_{36}H_{38}N_4O_7 \cdot 0.5\ H_2O$ (647.704): C 66.75, H 6.07, N 8.65; found: C 66.71, H 6.16, N 8.51.

Boc–(S)-Bin–Gly–Gly–(S)-Bin–Gly–OMe (**5**). Boc–Gly–Gly–(*S*)-Bin–Gly–OMe (**4**; 0.067 g, 0.11 mmol) was *N*-deprotected in CH_2Cl_2 (5 ml) and CF_3COOH (5 ml) as described above for Boc–Gly–(*S*)-Bin–Gly–OMe. To a soln. of the crude H–Gly–Gly–(*S*)-Bin–Gly–OMe (0.056 g, 0.11 mmol), Boc–(*S*)-Bin–OH (0.047 g, 0.11 mmol), and HOBt (0.021 g, 0.16 mmol) in THF (10 ml) and CH_2Cl_2 (7.5 ml), a soln. of EDC (0.024 g, 0.13 mmol) in CH_2Cl_2 (2.5 ml) was added. The mixture was magnetically stirred at r.t. for 48 h and evaporated. The residue was dissolved in AcOEt (150 ml) and the soln. extracted as described above for Boc–(*S*)-Bin–Ala–OMe. CC (silica gel, *B*) of the crude product followed by crystallization from CH_2Cl_2/hexane gave 0.076 g (74%) of pure **5**. Solid. M.p. 223°. $[\alpha]_{589}^{25} = -116$, $[\alpha]_{578}^{25} = -125$, $[\alpha]_{546}^{25} = -150$, $[\alpha]_{436}^{25} = -369$, $[\alpha]_{365}^{25} = -1361$ ($c = 0.2$, MeOH). R_f 0.40 (C). ^{1}H-NMR (Boc–Bin^1–Gly^2–Gly^3–Bin^4–Gly^5–OMe): 8.04 (br. *t*, NH (Gly^2 or Gly^3)); 7.97–7.01 (*m*, 24 arom. H); 7.20 (masked *d*, identified by 2D COSY, NH (Gly^5)); 7.17 (masked *d*, identified by 2D COSY, NH (Gly^3 or Gly^2)); 6.94 (*s*, NH (Bin^4)); 5.04 (*s*, NH (Bin^1)); 4.08–3.88 (*dd*, masked *dd*, $J = 16.1, 5.7$, 2 H–C(α) (Gly^5)); 3.87 (masked *d*, 2 H–C(α) (Gly^3 or Gly^2)); 3.86–3.70 (2 masked *dd*, $J \approx 17.8, 6.2$, 2 H–C(α) (Gly^2 or Gly^3)); 3.56 (*s*, MeO); 3.46–2.55 (*dd*, $J = 13.2$, 2 H–C(β) (Bin)); 3.11–2.20 (*dd*, $J \approx 13.2$, 2 H–C(β) (Bin)); 2.72 (*s*, 2 H–C(β) (Bin)); 1.35 (*s*, Boc). ^{13}C-NMR: 173.7, 172.9, 170.9, 170.5, 169.1 (C=O (Gly^1, Gly^2, Gly^3, Bin^1, Bin^2)); 156.0 (C=O (Boc)); 134.6–124.9 (arom. C); 81.4 (Boc); 70.6, 69.9 (C(α) (Bin^1, Bin^2)); 52.0 (MeO); 44.1, 43.7, 42.2 (C(α) (Gly^1, Gly^2, Gly^3)); 42.1, 41.3, 7.2, 36.9 (C(β), C(β') (Bin^1, Bin^2)); 28.2 (Boc). ESI-MS (pos.): 996 (100,

$[M+Na]^+$), 974 (31, $[M+H]^+$). Anal. calc. for $C_{60}H_{55}N_5O_8 \cdot H_2O$ (992.096): C 72.63, H 5.79, N 7.06; found: C 72.72, H 5.77, N 7.01.

Z–Aib–Aib–(S)-Bin–O^{t}Bu (**12**). A soln. of the oxazol-5(4*H*)-one from Z–Aib–Aib–OH [38] (0.336 g, 1.10 mmol) and H–(*S*)-Bin–O^{t}Bu [28] (0.368 g, 0.90 mmol) in MeCN (25 ml) was refluxed for 4 d and then evaporated. The residue was solubilized in AcOEt (150 ml). The soln. was extracted with 5% citric acid (2 × 50 ml), H_2O (100 ml), 5% $NaHCO_3$ soln. (2 × 50 ml), H_2O (2 × 100 ml), dried ($MgSO_4$), and evaporated. The crude product was submitted to CC (column 2 × 47 cm, silica gel, *B*) and then to prep. TLC (*A*): 0.500 g (78%) of pure **12**. Powder. M.p.129°. $[\alpha]_{589}^{25} = +89$, $[\alpha]_{578}^{25} = +92$, $[\alpha]_{546}^{25} = +103$, $[\alpha]_{436}^{25} = +136$, $[\alpha]_{365}^{25} = -167$ ($c = 0.7$, MeOH). R_f 0.30 (*B*). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–O^{t}Bu): 7.92–7.83 (*m*, 4 arom. H); 7.55 (*d*, $J = 8.3$, 1 arom. H); 7.45–7.06 (*m*, 13 H, arom. H, NH (Aib^2 or Bin)); 6.38 (*s*, NH (Bin or Aib^2)); 5.06 (*s*, NH (Aib^1)); 4.63–4.38 (*dd*, $J \approx 12.3$, CH_2 (Z)); 3.23–3.16 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 3.11–2.73 (*dd*, $J \approx 12.9$, 2 H–C(β) (Bin)); 1.57, 1.47 (2*s*, Me(β), Me(β') (Aib)); 1.49 (*s*, O^{t}Bu); 1.39, 1.32 (2*s*, Me(β), Me(β') (Aib)). ^{13}C-NMR: 173.2, 172.4, 170.8 (C=O (Aib^1, Aib^2, Bin)); 155.1 (C=O (Z)); 136.0–124.8 (arom. C); 80.9 (O^{t}Bu), 69.9 (C(α) (Bin)); 66.7 (CH_2 (Z)); 57.2, 56.7 (C(α) (Aib^1, Aib^2)); 41.1, 37.3 (C(β), C(β') (Bin)); 27.9 (O^{t}Bu); 25.8, 25.4, 25.2,25.0 (C(β), C(β') (Aib^1, Aib^2)). Anal. calc. for $C_{44}H_{47}N_3O_6 \cdot 0.5\ H_2O$ (722.848): C 73.11, H 6.69, N 5.81; found: C 73.25, H 6.76, N 5.69.

Z–Aib–Aib–(S)-Bin–OH. To a soln. of Z–Aib–Aib–(*S*)-Bin–O^{t}Bu (**12**; 0.481 g, 0.67 mmol) in CH_2Cl_2 (5 ml), CF_3COOH (5 ml) was added. The soln. was stirred at r.t. for 3 h and evaporated. The residue was solubilized in AcOEt (150 ml). The soln. was extracted with H_2O (2 × 100 ml), dried ($MgSO_4$), and evaporated. The residue was dissolved in MeOH and the soln. filtered and evaporated: 0.443 g (100%) of crude tripeptide as a solid, which was used in the next step without further purification. M.p. 205°. $[\alpha]_{589}^{25} = +100$, $[\alpha]_{578}^{25} = +103$, $[\alpha]_{546}^{25} = +116$, $[\alpha]_{436}^{25} = +160$, $[\alpha]_{365}^{25} = -129$ ($c = 0.2$, MeOH). R_f 0.30 (C). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–OH): 7.92–7.79 (*m*, 4 arom. H); 7.51–7.06 (*m*, 13 arom. H); 6.80, 6.78 (2*s*, 2 H, NH (Aib^2, Bin)); 6.28 (*s*, NH (Aib^1)); 4.35 (br. *m*, CH_2 (Z)); 3.30–3.14 (br. *dd*, $J \approx 13.6$, 2 H–C(β) (Bin)); 3.02–2.79 (br. *dd*, $J \approx 13.2$, 2 H–C(β) (Bin)); 1.50, 1.41 (2*s*, Me(β), Me(β') (Aib)); 1.41, 1.38 (2*s*, Me(β), Me(β') (Aib)). ^{13}C-NMR: 174.3, 173.6, 171.2 (C=O (Aib^1, Aib^2, Bin)); 155.9 (C=O (Z)); 135.7–125.1 (arom. C); 70.1 (C(α) (Bin)); 66.3 (CH_2 (Z)); 56.9, 56.7 (C(α) (Aib^1, Aib^2)); 40.8, 36.8 (C(β), (C(β') (Bin)); 26.3, 25.6, 23.8, 23.3 (C(β), C(β') (Aib^1, Aib^2). Anal. calc. for $C_{40}H_{39}N_3O_6 \cdot 1.5\ H_2O$ (684.760): C 70.15, H 6.18, N 6.14; found: C 70.01, H 6.16, N 6.02.

Oxazol-5(4H)-one from Z–Aib–Aib–(S)-Bin–OH. A soln. of Z–Aib–Aib–(*S*)-Bin–OH (0.433 g, 0.67 mmol) in Ac_2O (20 ml) was stirred at 115–120° for 40 min and then evaporated. The residue was repeatedly dissolved in toluene and the soln. evaporated: 0.430 g (100%) of the crude oxazolone as a solid, which was used in the next step without further purification. M.p. 163°. $[\alpha]_{589}^{25} = +127$, $[\alpha]_{578}^{25} = +128$, $[\alpha]_{546}^{25} = +143$, $[\alpha]_{436}^{25} = +172$, $[\alpha]_{365}^{25} = -313$ ($c = 0.2$, $CHCl_3$). R_f 0.65 (*B*), 0.80 (*C*). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–ox): 7.97–7.89 (*m*, 4 arom. H); 7.50–7.17 (*m*, 13 arom. H); 6.96 (br. *s*, NH (Aib^2)); 5.22 (*s*, NH (Aib^1)); 5.10 (*s*, CH_2 (Z)); 3.05–2.62 (*dd*, $J \approx 13.6$, 2 H–C(β) (Bin)); 2.97–2.65 (*dd*, $J \approx 13.3$, 2 H–C(β) (Bin)); 1.61, 1.59 (2*s*, Me(β), Me(β') (Aib)); 1.54, 1.52 (2*s*, Me(β), Me(β') (Aib)). Anal. calc. for $C_{40}H_{37}N_3O_5 \cdot 1.2\ H_2O$ (661.339): C 72.64, H 6.00, N 6.35; found: C 72.64, H 6.07, N 6.15.

Z–Aib–Aib–(S)-Bin–Aib–O^{t}Bu (**13**). HCl · H–Aib–O^{t}Bu [39] (0.315 g, 1.61 mmol) was stirred for a few min in Et_2O (75 ml) and 5% $NaHCO_3$ soln. (20 ml). The separated org. phase was dried ($MgSO_4$) and evaporated at 25°. A soln. of the resulting H–Aib–O^{t}Bu (0.220 g, 1.38 mmol) and the oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–OH (0.412 g, 0.65 mmol) in MeCN (15 ml) was refluxed for 24 h and then treated as described above for **12**. The crude product was submitted to CC (column 2.3 × 49 cm, silica gel, *B*): 0.427 g (83%) of pure **13**. Solid. M.p. 157°. $[\alpha]_{589}^{25} = +30$, $[\alpha]_{578}^{25} = +30$, $[\alpha]_{546}^{25} = +32$, $[\alpha]_{436}^{25} = +13$, $[\alpha]_{365}^{25} = -364$ ($c = 0.2$, MeOH). R_f 0.60 (*C*). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–Aib^3–O^{t}Bu): 7.87–7.77 (*m*, 4 arom. H); 7.58 (*d*, $J = 8.3$, 1 arom. H); 7.43–7.02 (*m*, 12 arom. H); 6.88, 6.86, 6.31 (3*s*, 3 NH (Aib^2, Aib^3, Bin)); 5.36 (*s*, NH (Aib^1)); 4.46–4.42 (*dd*, $J \approx 12.9$, CH_2 (Z)); 3.52–3.23 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 3.17–2.63 (*dd*, $J \approx 13.1$, 2 H–C(β) (Bin)); 1.52 (*s*, ^{t}BuO); 1.54, 1.51, 1.47, 1.41, 1.47, 1.41 (6*s*, 18 H, Me(β), Me(β) (Aib). ^{13}C-NMR: 174.1, 173.8, 172.6, 171.2 (C=O (Aib^1, Aib^2, Aib^3, Bin)); 155.4 (C=O (Z)); 136.4–124.7 (arom. C); 80.2 (^{t}BuO); 70.4 (C(α) (Bin)); 67.0 (CH_2 (Z)); 57.1, 56.8, 56.3 (C(α) (Aib^1, Aib^2, Aib^3)); 42.7, 34.7 (C(β), C(β') (Bin)); 27.9 (^{t}BuO); 27.5, 26.4, 25.7, 24.0, 23.7, 23.2 (C(β), C(β') (Aib^1, Aib^2, Aib^3)). Anal. calc. for $C_{48}H_{54}N_4O_7$ (798.944): C 72.16, H 6.81, N 7.01; found: C 72.24, H 6.96, N 6.92.

Z–Aib–Aib–(S)-Bin–Aib–OH. Z–Aib–Aib–(*S*)-Bin–Aib–O^{t}Bu (**13**; 0.422 g, 0.52 mmol) was *C*-deprotected in CF_3COOH (5 ml) and CH_2Cl_2 (5 ml) as described above for Z–Aib–Aib–(*S*)-Bin–OH: 0.379 g (96%) of crude tetrapeptide as a solid, which was used in the next step without further purification. M.p. 219°. $[\alpha]_{589}^{25} = +37$, $[\alpha]_{578}^{25} = +37$, $[\alpha]_{546}^{25} = +40$, $[\alpha]_{436}^{25} = +20$, $[\alpha]_{365}^{25} = -363$ ($c = 0.2$, MeOH). R_f 0.10 (*C*).

^{1}H-NMR (Z–Aib^1–Aib^2–Bin–Aib^3–OH): 7.89–7.69 (*m*, 6 arom. H); 7.41–7.05 (*m*, 11 arom. H); 7.01, 6.73, 6.70 (3*s*, 3 NH (Aib^2, Aib^3, Bin)); 6.11 (*s*, NH (Aib^1)); 4.08 (br. *m*, CH_2 (Z)); 3.26–3.04 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 2.87–2.82 (*dd*, $J \approx 13.4$, 2 H–C(β) (Bin)); 1.67, 1.52, 1.41, 1.30, 1.28, 1.27 (6*s*, 18 H, Me(β), Me(β') (Aib)). ^{13}C-NMR: 176.3, 175.5, 174.8, 172.8 (C=O (Aib^1, Aib^2, Aib^3, Bin)); 155.9 (C=O (Z)); 135.6–125.0 (arom. C); 70.0 (C(α), (Bin)); 66.3 (CH_2 (Z)); 57.2, 56.9, 56.7 (C(α) (Aib^1, Aib^2, Aib^3)); 42.2, 35.9 (C(β), C(β') (Bin)); 27.2, 26.1, 25.9, 24.2, 23.2, 22.6 (C(β), C(β') (Aib^1, Aib^2, Aib^3)). Anal. calc. for $C_{44}H_{46}N_4O_7 \cdot H_2O$ (760.856): C 69.45, H 6.36, N 7.36; found: C 69.41, H 6.38, N 7.21.

*Oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–Aib–OH.* Z–Aib–Aib–(*S*)-Bin–Aib–OH (0.372 g, 0.50 mmol) was treated with Ac_2O as described above for the oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–OH to give 0.359 g (99%) of crude oxazolone as a solid, which was used in the next step without further purification. M.p. 188°. $[\alpha]_{589}^{25} = +82$, $[\alpha]_{578}^{25} = +83$, $[\alpha]_{546}^{25} = +93$, $[\alpha]_{436}^{25} = +108$, $[\alpha]_{365}^{25} = -239$ ($c = 0.3$, MeOH). R_f 0.50 (*C*). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–Aib^3–ox): 7.91–7.86 (*m*, 3 arom. H); 7.81–7.66 (*dd*, $J = 8.3$, 2 arom. H); 7.46–7.10 (*m*, 12 arom. H); 7.09, 6.20 (2*s*, 2 NH (Aib^2, Bin)); 4.92 (*s*, NH (Aib^1)); 4.72–4.45 (*dd*, $J \approx 12.2$, CH_2 (Z)); 3.41–3.27 (*dd*, $J \approx 14.0$, 2 H–C(β) (Bin)); 3.02–2.95 (*dd*, $J \approx 13.1$, 2 H–C(β) (Bin)); 1.44, 1.39, 1.37, 1.36, 1.28, 1.26 (6*s*, 18 H, Me(β), Me(β') (Aib)). Anal. calc. for $C_{44}H_{44}N_4O_6 \cdot 0.5\ H_2O$ (733.832): C 72.01, H 6.18, N 7.63; found: C 72.34, H 6.29, N 7.49.

*Z–Aib–Aib–(*S*)-Bin–Aib–Aib–O^{t}Bu* (**14**). A soln. of H–Aib–O^{t}Bu (0.166 g, 1.04 mmol), obtained as described above for **13**, and the oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–Aib–OH (0.351 g, 0.48 mmol) in MeCN (20 ml), was refluxed for 40 h and then treated as described above for Z–Aib–Aib–(*S*)-Bin–Aib–O^{t}Bu. CC (column 2 × 51 cm, silica gel, *B*) of the crude product gave 0.346 g (81%) of pure **14**. Solid. M.p. 252°. $[\alpha]_{589}^{25} = +6$, $[\alpha]_{578}^{25} = +5$, $[\alpha]_{546}^{25} = +3$, $[\alpha]_{436}^{25} = -38$, $[\alpha]_{365}^{25} = -400$ ($c = 0.2$, MeOH). R_f 0.65 (*C*). ^{1}H-NMR (Z–Aib^1–Aib^2–Bin–Aib^3–Aib^4–O^{t}Bu): 7.87–7.73 (*m*, 4 arom. H); 7.40–7.07 (*m*, 13 arom. H); 7.52, 6.72, 6.69, 6.47 (4*s*, 4 NH (Aib^2, Aib^3, Aib^4, Bin)); 6.21 (*s*, NH (Aib^1)); 4.06–4.02 (*dd*, $J \approx 12.3$, CH_2 (Z)); 3.28–3.13 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 2.86–2.80 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 1.42 (*s*, ^{t}BuO); 1.66, 1.53, 1.51, 1.48, 1.47, 1.42 (masked), 1.27, 1.21 (8*s*, 24 H, Me(β), Me(β') (Aib)). ^{13}C-NMR: 174.3, 174.2, 174.0, 173.7, 171.0 (C=O (Aib^1, Aib^2, Aib^3, Aib^4, Bin)); 155.8 (C=O (Z)); 136.2–124.9 (arom. C); 79.7 (^{t}BuO); 70.2 (C(α) (Bin)); 66.5 CH_2 (Z)); 57.0, 56.6, 56.5, 55.9 (C(α) (Aib^1, Aib^2, Aib^3, Aib^4)); 42.5, 35.3 (C(β), C(β') (Bin)); 27.8 (^{t}BuO); 27.7, 26.5, 25.9, 24.2, 23.2, 23.0, 22.74, 22.71 (C(β), C(β') (Aib^1, Aib^2, Aib^3, Aib^4)). Anal. calc. for $C_{52}H_{61}N_5O_8 \cdot 0.5\ H_2O$ (893.056): C 69.93, H 7.00, N 7.84; found: C 69.92, H 7.05, N 7.82.

*Z–(*S*)-Bin–Aib–O^{t}Bu.* To a soln. of Z–(*S*)-Bin–OH (0.243 g, 0.5 mmol) and Et_3N (0.084 ml, 0.6 mmol) in toluene (2 ml) cooled to −5°, a soln. of Piv-Cl (0.074 ml, 0.6 mmol) was added. The resulting suspension was stirred at −5° for 1.5 h, then at r.t. for 2 h, and evaporated at 30°. To the solid mixture (containing crude Z–(*S*)-Bin–OPiv), a soln. of H–Aib–O^{t}Bu (0.225 g, 1.4 mmol; obtained as described above for **13**) in toluene (10 ml) was added. The mixture was stirred at 60° overnight, then evaporated and treated as described above for **13**. The crude product was submitted to CC (column 1.5 × 41 cm, silica gel, *A*, then *C*): 0.097 g (40%) of recovered pure Z–(*S*)-Bin–OH and 0.171 g (54%) of pure dipeptide. Solid. M.p. 119°. $[\alpha]_{589}^{25} = -4$, $[\alpha]_{578}^{25} = -5$, $[\alpha]_{546}^{25} = -10$, $[\alpha]_{436}^{25} = -92$, $[\alpha]_{365}^{25} = -687$ ($c = 0.2$, MeOH). R_f 0.85 (*B*). ^{1}H-NMR: 7.94–7.83 (*m*, 4 arom. H); 7.55 (*d*, $J = 8.4$, 1 arom. H); 7.47–7.19 (*m*, 13 H, arom. H, NH (Aib)); 5.24–5.09 (*d* · br. *d*, $J \approx 12.1$, CH_2 (Z)); 4.93 (*s*, NH (Bin)); 3.44–2.32 (*dd*, $J \approx 12.9$, 2 H–C(β) (Bin)); 3.31–3.06 (br. *dd*, $J \approx 13.2$, 2 H–C(β) (Bin)); 1.55, 1.50 (2*s*, Me(β), Me(β') (Aib)); 1.43 (*s*, ^{t}BuO). ^{13}C-NMR: 173.9, 170.6 (C=O (Aib, Bin)); 155.1 (C=O (Z)); 136.2–125.1 (arom. C); 81.3 (^{t}BuO); 70.6 (C(α) (Bin)); 66.7 CH_2 (Z)); 56.8 (C(α) (Aib)); 42.2, 37.1 (C(β), C(β') (Bin)); 27.7 (^{t}BuO); 24.2, 24.0 (C(β), C(β') (Aib)). Anal. calc. for $C_{40}H_{40}N_2O_5$ (628.736): C 76.41, H 6.41, N 4.46; found: C 76.14, H 6.53, N 4.27.

*Z–(*S*)-Bin–Aib–OH.* Z–(*S*)-Bin–Aib–O^{t}Bu (0.162 g, 0.26 mmol) was *C*-deprotected in CF_3COOH (5 ml) and CH_2Cl_2 (5 ml) as described above for the synthesis of Z–Aib–Aib–(*S*)-Bin–OH to give 0.138 g (93%) of crude dipeptide as a solid, which was used in the next step without further purification. M.p. 193°. $[\alpha]_{589}^{25} = -11$, $[\alpha]_{578}^{25} = -14$, $[\alpha]_{546}^{25} = -21$, $[\alpha]_{436}^{25} = -116$, $[\alpha]_{365}^{25} = -762$ ($c = 0.2$, MeOH). R_f 0.10 (*B*). Anal. calc. for $C_{36}H_{32}N_2O_5 \cdot H_2O$ (590.648): C 73.20, H 5.80, N 4.74; found: C 73.59, H 5.94, N 4.55.

*Oxazol-5(4*H*)-one from Z–(*S*)-Bin–Aib–OH.* Z–(*S*)-Bin–Aib–OH (0.130 g, 0.23 mmol) was treated with Ac_2O as described above for oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–OH to give 0.125 g (99%) of the crude oxazolone as a glassy solid, which was used in the next step without further purification. $[\alpha]_{589}^{25} = +10$, $[\alpha]_{578}^{25} = +5$, $[\alpha]_{546}^{25} = +4$, $[\alpha]_{436}^{25} = -53$, $[\alpha]_{365}^{25} = -531$ ($c = 0.2$, $CHCl_3$). R_f 0.80 (*B*). ^{1}H-NMR: 7.96–7.83 (*m*, 4 arom. H); 7.62–7.23 (*m*, 13 arom. H); 5.20–5.10 (*d* · br. *d*, $J \approx 12.1$, CH_2 (Z)); 4.89 (*s*, NH (Bin)); 3.31–3.18 (br. *dd*, $J \approx 14.0$, 2 H–C(β) (Bin)); 3.25–2.48 (*dd*, $J \approx 12.9$, 2 H–C(β) (Bin)); 1.38, 1.26 (2*s*, Me(β), Me(β'), Aib). Anal. calc. for $C_{36}H_{30}N_2O_4 \cdot 2\ H_2O$ (590.648): C 73.20, H 5.80, N 4.74; found: C 73.28, H 5.89, N 4.62.

Z–(S)-Bin–Aib–Aib–O^{t}Bu (**9**). A soln. of H–Aib–O^{t}Bu (0.166 g, 1.04 mmol), obtained as described above for the synthesis of Z–Aib–Aib–(*S*)-Bin–Aib–O^{t}Bu, and the oxazol-5-(4*H*)-one from Z–(*S*)-Bin–Aib–OH (0.119 g, 0.21 mmol) in MeCN (30 ml), was refluxed for 72 h and then treated as described above for Z–Aib–Aib–(*S*)-Bin–O^{t}Bu. CC (column 2 × 51 cm, silica gel, *B*) of the crude product gave 0.139 g (91%) of pure **9**. Solid. M.p. 130°. $[\alpha]_{589}^{25} = -58$, $[\alpha]_{578}^{25} = -62$, $[\alpha]_{546}^{25} = -75$, $[\alpha]_{436}^{25} = -200$, $[\alpha]_{365}^{25} = -801$ (*c* = 0.2, MeOH). R_f 0.50 (*B*). ^{1}H-NMR (Z–Bin–Aib^1–Aib^2–O^{t}Bu): 7.93–7.83 (*m*, 4 arom. H); 7.44–7.19 (*m*, 14 H, arom. H, NH (Aib)); 6.48 (*s*, NH (Aib)); 5.23 (*s*, NH (Bin)); 5.21–5.10 (*dd*, *J* ≈ 12.2, CH$_2$ (Z)); 3.17–2.31 (*dd*, *J* ≈ 12.8, 2 H–C(β) (Bin)); 3.13–3.08 (*dd*, *J* ≈ 14.0, 2 H–C(β) (Bin)); 1.51,1.49, 1.47, 1.44 (4*s*, 12 H, Me(β), Me(β') (Aib^1, Aib^2); 1.45 (*s*, ^{t}BuO). ^{13}C-NMR: 173.6, 172.6, 170.3 (C=O (Aib^1, Aib^2, Bin)); 155.5 (C=O (Z)); 136.0–125.2 (arom. C); 80.3 (^{t}BuO); 70.4 (C(α) (Bin)); 67.0 CH$_2$ (Z)); 56.7, 56.2 (C(α) (Aib^1, Aib^2)); 42.6, 36.9 (C(β), C(β') (Bin)); 27.8 (^{t}BuO); 25.7, 24.9, 24.7, 24.1 (C(β), C(β') (Aib^1, Aib^2)). Anal. calc. for $C_{44}H_{47}N_3O_6$ (713.840): C 74.03, H 6.63, N 5.89; found: C 74.26, H 6.68, N 5.81.

Z–(S)-Bin–Aib–Aib–OH. Z–(*S*)-Bin–Aib–Aib–O^{t}Bu (**9**; 0.134 g, 0.19 mmol) was *C*-deprotected in CF$_3$COOH (5 ml) and CH$_2$Cl$_2$ (5 ml) as described above for Z–Aib–Aib–(*S*)-Bin–OH to give 0.118 g (96%) of crude tripeptide as a solid, which was used in the next step without further purification. M.p. 219°. $[\alpha]_{589}^{25} = -46$, $[\alpha]_{578}^{25} = -49$, $[\alpha]_{546}^{25} = -62$, $[\alpha]_{436}^{25} = -180$, $[\alpha]_{365}^{25} = -790$ (*c* = 0.2, MeOH). R_f 0.05 (*B*). Anal. calc. for $C_{40}H_{39}N_3O_6 \cdot 2.5\ H_2O$ (590.648): C 68.36, H 6.31, N 5.98; found: C 68.71, H 6.47, N 5.85.

*Oxazol-5(4*H*)-one from Z–(S)-Bin–Aib–Aib–OH.* Z–(*S*)-Bin–Aib–Aib–OH (0.112 g, 0.17 mmol) was treated with Ac$_2$O as described above for the oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–OH to give 0.109 g (100%) of crude oxazolone as a powder, which was used in the next step without further purification. M.p. 140°. $[\alpha]_{589}^{25} = +44$, $[\alpha]_{578}^{25} = +43$, $[\alpha]_{546}^{25} = +45$, $[\alpha]_{436}^{25} = +10$, $[\alpha]_{365}^{25} = -469$ (*c* = 0.2, CHCl$_3$). R_f = 0.70 (*B*). ^{1}H-NMR (Z–Bin–Aib^1–Aib^2–ox): 7.94–7.82 (*m*, 4 arom. H); 7.54–7.19 (*m*, 13 arom. H); 6.83 (br. *s*, NH (Aib^1)); 5.22–5.11 (*d* · br. *d*, *J* ≈ 12.0, CH$_2$ (Z)); 4.87 (*s*, NH (Bin)); 3.33–2.35 (br. *dd*, *J* ≈ 12.9, 2 H–C(β) (Bin)); 3.20–3.10 (br. *dd*, *J* ≈ 13.6, 2 H–C(β) (Bin)); 1.54, 1.46, 1.42, 1.26 (4*s*, 12 H, Me(β), Me(β') (Aib^1, Aib^2)). Anal. calc. for $C_{40}H_{37}N_3O_5$ (675.752): C 71.09, H 6.12, N 6.22; found: C 71.01, H 6.21, N 6.02.

Z–(S)-Bin–Aib–Aib–(S)-Bin–O^{t}Bu (**10**). A soln. of the oxazol-5(4*H*)-one from Z–(*S*)-Bin–Aib–Aib–OH (0.104 g, 0.16 mmol) and H–(*S*)-Bin–O^{t}Bu (0.082 g, 0.2 mmol) in MeCN (30 ml) was refluxed for 21 d, then treated as described above for **12**. The crude product was submitted to CC (column 1.5 × 43 cm, silica gel, *A*, then *B* and *C*), followed by prep. TLC (*B*): 0.083 g (49%) of pure **10**. Solid. M.p. 228°. $[\alpha]_{589}^{25} = -131$, $[\alpha]_{578}^{25} = -140$, $[\alpha]_{546}^{25} = -169$, $[\alpha]_{436}^{25} = -413$, $[\alpha]_{365}^{25} = -1472$ (*c* = 0.2, MeOH). R_f 0.40 (*B*). ^{1}H-NMR (Z–Bin^1–Aib^2–Aib^3–Bin^4–O^{t}Bu): 7.93–6.95 (*m*, 31 H, 29 arom. H, 2 NH (Aib or Bin^4)); 6.12 (*s*, NH (Aib or Bin^4)); 5.25–5.06 (*dd*, *J* ≈ 12.1, CH$_2$ (Z)); 5.01 (*s*, NH (Bin^1)); 3.29–3.14 (*dd*, *J* ≈ 13.9, 2 H–C(β) (Bin)); 3.13–2.86 (*dd*, *J* ≈ 13.1, 2 H–C(β) (Bin)); 2.96–2.08 (*dd*, *J* ≈ 13.1, 2 H–C(β) (Bin)); 2.59 (*s*, 2 H–C(β) (Bin)); 1.65, 1.57, 1.34, 1.14 (4*s*, 12 H, Me(β), Me(β') (Aib^2, Aib^3)); 1.51 (*s*, ^{t}BuO). ^{13}C-NMR: 173.9, 171.9, 171.3 (C=O (Aib^2, Aib^3, Bin^1, Bin^4)); 155.6 (C=O (Z)); 136.3–124.4 (arom. C); 80.6 (^{t}BuO); 70.0, 69.9 (C(α) (Bin^1, Bin^4)); 67.4 CH$_2$ (Z)); 57.0, 56.8 (C(α) (Aib^2, Aib^3)); 42.1, 41.2, 37.8, 37.1 (C(β), C(β') (Bin^1, Bin^4)); 28.0 (^{t}BuO); 26.6, 26.0, 24.7, 24.5 (C(β), C(β') (Aib^2, Aib^3)). Anal. calc. for $C_{68}H_{64}N_4O_7 \cdot 0.5\ H_2O$ (1058.232): C 77.17, H 6.19, N 5.29; found: C 77.17, H 6.29, N 5.11.

Z–(S)-Bin–Aib–Aib–(S)-Bin–OH. Z–(*S*)-Bin–Aib–Aib–(*S*)-Bin–O^{t}Bu (**10**; 0.076 g, 0.07 mmol) was *C*-deprotected in CF$_3$COOH (5 ml) and CH$_2$Cl$_2$ (5 ml) as described above for Z–Aib–Aib–(*S*)-Bin–OH to give 0.071 g (99%) of crude tetrapeptide as a solid, which was used in the next step without further purification. M.p. 280°. $[\alpha]_{589}^{25} = -130$, $[\alpha]_{578}^{25} = -139$, $[\alpha]_{546}^{25} = -167$, $[\alpha]_{436}^{25} = -407$, $[\alpha]_{365}^{25} = -1460$ (*c* = 0.2, MeOH). R_f 0.25 (*B*), 0.40 (*C*). Anal. calc. for $C_{64}H_{56}N_4O_7 \cdot H_2O$ (1011.136): C 76.01, H 5.78, N 5.54; found: C 76.03, H 5.92, N 5.38.

*Oxazol-5(4*H*)-one from Z–(S)-Bin–Aib–Aib–(S)-Bin–OH.* Z–(*S*)-Bin–Aib–Aib–(*S*)-Bin–OH (0.067 g, 0.07 mmol) was treated with Ac$_2$O as described above for the oxazol-5(4*H*)-one from Z–Aib–Aib–(*S*)-Bin–OH to give 0.066 g (100%) of crude oxazolone as a powder, which was used in the next step without further purification. M.p. 169°. $[\alpha]_{589}^{25} = +62$, $[\alpha]_{578}^{25} = +59$, $[\alpha]_{546}^{25} = +63$, $[\alpha]_{436}^{25} = +7$, $[\alpha]_{365}^{25} = -698$ (*c* = 0.2, CHCl$_3$). R_f 0.85 (*B*), 0.90 (*C*). Anal. calc. for $C_{64}H_{54}N_4O_6 \cdot 3\ H_2O$ (1029.152): C 74.69, H 5.88, N 5.44; found: C 75.01, H 6.29, N 5.07.

Z–(S)-Bin–Aib–Aib–(S)-Bin–Aib–O^{t}Bu (**11**). A soln. of H–Aib–O^{t}Bu (0.067 g, 0.42 mmol, obtained as described above for the synthesis of **13**) and the oxazol-5(4*H*)-one from Z–(*S*)-Bin–Aib–Aib–(*S*)-Bin–OH (0.062 g, 0.06 mmol) in MeCN (10 ml) was refluxed for 72 h and then treated as described above for **12**. The crude product was submitted to CC (column 1.5 × 40 cm, silica gel, *B*), then to prep. TLC (*A*); 0.056 g (78%) of pure **11**. Solid. M.p. 213°. $[\alpha]_{589}^{25} = -183$, $[\alpha]_{578}^{25} = -194$, $[\alpha]_{546}^{25} = -230$, $[\alpha]_{436}^{25} = -532$, $[\alpha]_{365}^{25} = -1674$ (*c* = 0.2, MeOH). R_f 0.40 (*B*). ^{1}H-NMR (Z–Bin^1–Aib^2–Aib^3–Bin^4–Aib^5–O^{t}Bu): 7.93–6.47 (*m*, 32 H,

29 arom. H, 3 NH (Aib or Bin[4])); 6.37 (*s*, NH (Aib or Bin[4])); 5.10 (*s*, CH_2 (Z)); 5.03 (*s*, NH (Bin[1])); 3.31–2.97 (*dd*, $J \approx 13.5$, 2 H–C(β) (Bin)); 2.72–2.17 (*dd*, $J \approx 13.2$, 2 H–C(β) (Bin)); 2.66–2.18 (*dd*, $J \approx 13.8$, 2 H–C(β) (Bin)); 2.00–1.91 (*dd*, $J \approx 14.0$, 2 H–C(β) (Bin)); 1.53 (*s*, ^{t}BuO); 1.53 (masked), 1.51, 1.50, 1.49, 1.41, 1.24 (6*s*, 18 h, Me(β), Me(β') (Aib[2], Aib[3], Aib[5])). ^{13}C-NMR: 174.2, 173.6, 173.4, 171.3, 171.0 (C=O (Aib[2], Aib[3], Aib[5], Bin[1], Bin[4])); 155.6 (C=O (Z)); 136.1–124.2 (arom. C); 80.1 (^{t}BuO); 70.0, 69.9 (Cα) (Bin[1], Bin[4])); 67.4 (CH_2 (Z)); 56.9, 56.8, 56.1 (C(α) (Aib[2], Aib[3], Aib[5])); 42.6, 42.3, 35.5, 34.4 (C(β), C(β') (Bin[1], Bin[4])); 27.9 (^{t}BuO); 27.3, 26.7, 25.8, 23.9, 23.3 (C(β), C(β') (Aib[2], Aib[3], Aib[5])). Anal. calc. for $C_{72}H_{71}N_5O_8 \cdot H_2O$ (1152.344): C 75.04, H 6.39, N 6.08; found: C 74.71, H 6.55, N 5.91.

X-Ray Diffraction. The compounds investigated, $H^+ \cdot H-(S)$-Bin$-O^-$ and Bz–(*R*)-Bin–Phe–NHChx, are chiral and accordingly they crystallize in a non-centrosymmetric space group, $P2_1$ for the free amino acid and $P2_12_12_1$ for the terminally blocked dipeptide. For these two structures, data were collected on an *Enraf-Nonius CAD-4* diffractometer with graphite-monochromated CuK_α ($\lambda = 1.54184$ Å) radiation. Lattice parameters were obtained by accurate centring of 25 standard reflections. Both structures were solved with the program SHELXS 86 [71]. Refinement was carried out with SHELXL 93 [72] for the free amino acid and with SHELXL 97 [73] for the dipeptide. In the structure of the free amino acid, the NH H-atoms were located on a difference-*Fourier* map, while H-atoms bound to C-atoms were placed in an ideal position. All H-atoms of the free amino acid were refined with individual isotropic thermal parameters. In the structure of the dipeptide, all H-atoms were calculated at idealized positions and refined as riding, with U_{iso} set equal to 1.2 times the U_{eq} of the

Table 3. *Crystallographic Data and Details of Structure Determinations for $H^+ \cdot H-$(S)-Bin$-O^-$ Methanol Bis-Solvate and Bz–(R)-Bin–Phe–NHChx* (**1′**) *Ethyl Acetate Bis-Solvate*

	$H^+ \cdot H-(S)$-Bin$-O^-$	Bz–(*R*)-Bin–Phe–NHChx (**1′**)
Empirical formula	$C_{26}H_{27}NO_4$	$C_{54}H_{59}N_3O_7$
M_r	417.5	862.0
Crystal system	monoclinic	orthorhombic
Space group	$P2_1$	$P2_12_12_1$
a [Å]	12.705(3)	11.225(4)
b [Å]	6.411(2)	16.717(10)
c [Å]	13.411(2)	26.156(6)
β [°]	95.38(1)	90
V [Å^3]	1130.6(5)	4908(4)
Z	2	4
D_x [Mg·m^{-3}]	1.226	1.167
μ [mm^{-1}]	0.662	0.615
Radiation	CuK_α ($\lambda = 1.5418$ Å)	CuK_α ($\lambda = 1.5418$ Å)
Crystal size [mm]	$0.8 \times 0.5 \times 0.3$	$0.5 \times 0.1 \times 0.1$
Mode of scan	$\omega - 2\theta$	$\omega - 2\theta$
F (000)	444	1840
θ Range [°]	3.2–68.0	4.7–60.0
Index ranges	$-15 \le h \le 15$	$-12 \le h \le 12$
	$-6 \le k \le 7$	$0 \le k \le 17$
	$0 \le l \le 16$	$0 \le l \le 29$
Reflections collected	4634	7926
Independent reflections	2259 (R(int) = 0.062)	6794 (R(int) = 0.058)
Absorption correction	none	none
Refinement method	full-matrix least squares on F^2	full-matrix least squares on F^2
Data/restraints/parameters	2244/1/361	6794/43/583
Goodness-of-fit on F^2	1.096	1.019
Final *R* indices ($I > 2\sigma(I)$)	$R_1 = 0.0562$	$R_1 = 0.0699$
	$wR_2 = 0.1309$	$wR_2 = 0.1812$
R Indices (all data)	$R_1 = 0.0828$	$R_1 = 0.1465$
	$wR_2 = 0.1640$	$wR_2 = 0.2223$
Largest difference peak and hole [e·Å^{-3}]	0.327/−0.438	0.197/−0.224

parent atom. The high displacement parameters observed for the terminal phenyl and cyclohexyl groups in the dipeptide indicated a low accuracy in the determination of the positions of their atoms. Crystallographic data and details of the structure determinations are given in *Table 3*. Atomic coordinates, bond distances and angles, and anisotropic displacement parameters have been deposited with the *Cambridge Crystallographic Data Centre* (CCDC) as deposition No. CCDC 150266 and 150267. Copies of the data can be obtained, free of charge, on application to the CCDC, 12 Union Road, Cambridge CB21EZ, UK (fax: +44 (1223)336033; e-mail: deposit@ccdc.cam.ac.uk).

F. F., *M. C.*, and *C. T.* gratefully acknowledge MURST, the Ministry of University and Scientific and Technological Research, and the *National Research Council* (CNR) of Italy for the continuous support to this research.

REFERENCES

[1] A. F. Spatola, in 'Chemistry and Biochemistry of Amino Acids, Peptides and Proteins', Ed. B. Weinstein, Dekker, New York, 1983, Vol. 7, p. 267.
[2] V. J. Hruby, F. Al-Obeidi, W. Kazmierski, *Biochem. J.* **1990**, *268*, 249.
[3] J. Rizo, L. M. Gierasch, *Annu. Rev. Biochem.* **1992**, *61*, 387.
[4] R. M. J. Liskamp, *Recl. Trav. Chim. Pays-Bas* **1994**, *113*, 1.
[5] J. Gante, *Angew. Chem., Int. Ed.* **1994**, *33*, 1699.
[6] S. Hanessian, G. McNaughton-Smith, H. G. Lombart, W. D. Lubell, *Tetrahedron* **1997**, *53*, 12789.
[7] C. Toniolo, *Int. J. Pept. Protein Res.* **1990**, *35*, 287.
[8] C. Toniolo, E. Benedetti, *Macromolecules* **1991**, *24*, 4004.
[9] G. Valle, M. Crisma, C. Toniolo, Sudhanand, R. Balaji Rao, M. Sukumar, P. Balaram, *Int. J. Pept. Protein Res.* **1991**, *38*, 511.
[10] C. Toniolo, M. Crisma, F. Formaggio, E. Benedetti, A. Santini, R. Iacovino, M. Saviano, B. Di Blasio, C. Pedone, J. Kamphuis, *Biopolymers* **1996**, *40*, 519.
[11] E. Benedetti, B. Di Blasio, R. Iacovino, V. Menchise, M. Saviano, C. Pedone, G. M. Bonora, A. Ettorre, L. Graci, F. Formaggio, M. Crisma, G. Valle, C. Toniolo, *J. Chem. Soc., Perkin Trans. 2* **1997**, 2023.
[12] D. Obrecht, C. Spiegler, P. Schönholzer, K. Müller, H. Heimgartner, F. Stierli, *Helv. Chim. Acta* **1992**, *75*, 1666.
[13] I. L. Karle, P. Balaram, *Biochemistry* **1990**, *29*, 6747.
[14] C. Toniolo, M. Crisma, F. Formaggio, G. Valle, G. Cavicchioni, G. Précigoux, A. Aubry, J. Kamphuis, *Biopolymers* **1993**, *33*, 1061.
[15] B. Jaun, M. Tanaka, P. Seiler, F. N. M. Kühnle, C. Braun, D. Seebach, *Liebigs Ann. Chem.* **1997**, 1697.
[16] K. Nebel, E. Altmann, M. Mutter, R. Bardi, A. M. Piazzesi, M. Crisma, G. M. Bonora, C. Toniolo, *Biopolymers* **1991**, *31*, 1135.
[17] C. M. Venkatachalam, *Biopolymers* **1968**, *6*, 1425.
[18] C. Toniolo, *C.R.C. Crit. Rev. Biochem.* **1980**, *9*, 1.
[19] G. D. Rose, L. M. Gierasch, J. P. Smith, *Adv. Protein Chem.* **1985**, *37*, 1.
[20] C. Toniolo, E. Benedetti, *Trends Biochem. Sci.* **1991**, *16*, 350.
[21] L. Pu, *Chem. Rev.* **1998**, *98*, 2405.
[22] L. Pu, *Tetrahedron: Asymmetry* **1998**, *9*, 1457.
[23] M. Sutala, *Enantiomer* **1999**, *4*, 243.
[24] K. Nozaki, T. Terakawa, H. Takaya, T. Hiyamo, *Tetrahedron: Asymmetry* **1997**, *8*, 3431.
[25] J. K. Whitesell, *Chem. Rev.* **1989**, *89*, 1581.
[26] R. Noyori, H. Takaya, *Acc. Chem. Res.* **1990**, *23*, 345.
[27] J.-P. Mazaleyrat, A. Gaucher, M. Wakselman, L. Tchertanov, J. Guilhem, *Tetrahedron Lett.* **1996**, *37*, 2971.
[28] J.-P. Mazaleyrat, A. Gaucher, J. Šavrda, M. Wakselman, *Tetrahedron: Asymmetry* **1997**, *8*, 619.
[29] J.-P. Mazaleyrat, A. Boutboul, Y. Lebars, A. Gaucher, M. Wakselman, *Tetrahedron: Asymmetry* **1998**, *9*, 2701.
[30] J.-P. Mazaleyrat, A. Gaucher, M. Wakselman, C. Toniolo, M. Crisma, F. Formaggio, in 'Peptides 1996', Eds. R. Ramage and R. Epton, Mayflower Scientific, Kingswinford, U.K., 1998, p. 623.
[31] C. Toniolo, F. Formaggio, M. Crisma, J.-P. Mazaleyrat, M. Wakselman, C. George, J. R. Deschamps, J. L. Flippen-Anderson, B. Pispisa, M. Venanzi, A. Palleschi, *Chem. – Eur. J.* **1999**, *5*, 2254.

[32] M. J. Burk, J. R. Lee, J. P. Martinez, *J. Am. Chem. Soc.* **1994**, *116*, 10847.
[33] L. Ridvan, N. Abdallah, R. Holakovsky, M. Tichy, J. Zavada, *Tetrahedron: Asymmetry* **1996**, *7*, 231.
[34] J.-P. Mazaleyrat, A. Gaucher, Y. Goubard, J. Savrda, M. Wakselman, *Tetrahedron Lett.* **1997**, *38*, 2091.
[35] J.-P. Mazaleyrat, Y. Goubard, M. V. Azzini, M. Wakselman, M. Crisma, F. Formaggio, C. Toniolo, in 'Peptides 2000', Eds. J. Martinez and R. Epton, Mayflower Scientific, Kingswinford, U.K., in press.
[36] A. Peter, G. Török, J.-P. Mazaleyrat, M. Wakselman, *J. Chromatogr.* **1997**, *790*, 41.
[37] W. König, R. Geiger, *Chem. Ber.* **1970**, *103*, 788.
[38] M. T. Leplawy, D. S. Jones, G. W. Kenner, R. C. Sheppard, *Tetrahedron* **1960**, *11*, 39.
[39] D. S. Jones, G. W. Kenner, J. Preston, R. C. Sheppard, *J. Chem. Soc.* **1965**, 6227.
[40] S. F. Mason, R. H. Seal, D. R. Roberts, *Tetrahedron* **1974**, *30*, 1671.
[41] N. Harada, K. Nakanishi, 'Circular Dichroic Spectroscopy. Exciton Coupling in Organic Stereochemistry', Oxford University Press, Oxford, 1983.
[42] L. Bi Bari, G. Pescitelli, P. Salvadori, *J. Am. Chem. Soc.* **1999**, *121*, 7998.
[43] S. Mizushima, T. Shimanouchi, M. Tsuboi, R. Souda, *J. Am. Chem. Soc.* **1952**, *74*, 270.
[44] G. M. Bonora, C. Mapelli, C. Toniolo, R. R. Wilkening, E. S. Stevens, *Int. J. Biol. Macromol.* **1984**, *6*, 179.
[45] D. F. Kennedy, M. Crisma, C. Toniolo, D. Chapman, *Biochemistry* **1991**, *30*, 6541.
[46] C. Toniolo, E. Benedetti, in 'Molecular Conformation and Biological Interactions', Eds. P. Balaram and S. Ramaseshan, Indian Academy of Sciences, Bangalore, India, 1991, p. 511.
[47] K. D. Kopple, M. Ohnishi, *Biochemistry* **1969**, *8*, 4087.
[48] K. D. Kopple, T. J. Schamper, *J. Am. Chem. Soc.* **1972**, *94*, 3644.
[49] P. Chakrabarti, J. D. Dunitz, *Helv. Chim. Acta* **1982**, *65*, 1555.
[50] E. Benedetti, in 'Chemistry and Biochemistry of Amino Acids, Peptides and Proteins', Ed. B. Weinstein, Dekker, New York, 1982, Vol. 6, p. 105.
[51] T. Ashida, Y. Tsunogae, I. Tanaka, T. Yamane, *Acta Crystallogr., Sect. B* **1987**, *43*, 212.
[52] IUPAC-IUB Commission on Biochemical Nomenclature, *J. Mol. Biol.* **1970**, *52*, 1.
[53] C. Ramakrishnan, N. Prasad, *Int. J. Protein Res.* **1971**, *3*, 209.
[54] R. Taylor, O. Kennard, W. Versichel, *Acta Crystallogr., Sect. B* **1984**, *40*, 280.
[55] C. H. Görbitz, *Acta Crystallogr., Sect. B* **1989**, *45*, 390.
[56] E. Benedetti, G. Morelli, G. Némethy, H. A. Scheraga, *Int. J. Pept. Protein Res.* **1983**, *22*, 1.
[57] Y. Umezawa, S. Tsuboyama, H. Takahashi, J. Uzawa, M. Nishio, *Bioorg. Med. Chem.* **1999**, *7*, 2021.
[58] J. B. Hendrickson, *J. Am. Chem. Soc.* **1967**, *89*, 7036.
[59] M. Bixon, S. Lifson, *Tetrahedron* **1967**, *23*, 769.
[60] D. F. Bocian, H. M. Pickett, T. C. Round, H. L. Strauss, *J. Am. Chem. Soc.* **1975**, *97*, 687.
[61] D. M. Ferguson, D. J. Raber, *J. Am. Chem. Soc.* **1989**, *111*, 4371.
[62] F. H. Allen, J. A. K. Howard, N. A. Pitchford, *Acta Crystallogr., Sect. B* **1993**, *49*, 910.
[63] D. Cremer, J. A. Pople, *J. Am. Chem. Soc.* **1975**, *97*, 1354.
[64] Cambridge Structural Data Base (CSD), Cambridge, U.K., October, 1996.
[65] I. D. Brown, *Acta Crystallogr., Sect. A* **1976**, *32*, 24.
[66] J. Mitra, C. Ramakrishnan, *Int. J. Pept. Protein Res.* **1977**, *9*, 27.
[67] M. Goodman, C. Toniolo, P. Pallai, in 'Forum Peptides', Eds. B. Castro and J. Martinez, Dohr, Nancy, 1985, p. 146.
[68] G. D. Fasman, 'Poly-α-Amino Acids: Protein Models for Conformational Studies', Dekker, New York, 1967.
[69] F. Formaggio, M. Crisma, C. Toniolo, L. Tchertanov, J. Guilhem, J.-P. Mazaleyrat, A. Gaucher, M. Wakselman, *Tetrahedron* **2000**, *56*, 8721.
[70] F. Formaggio, M. Crisma, C. Toniolo, J.-P. Mazaleyrat, M. Wakselman, in 'Peptides 1998', Eds. S. Bajusz and F. Hudecz, Akadémiai Kiadó, Budapest, 1999, p. 352.
[71] G. M. Sheldrick, 'SHELX 86, Program for the Solution of Crystal Structures', University of Göttingen, Germany, 1986.
[72] G. M. Sheldrick, 'SHELXL 93, Program for the Refinement of Crystal Structures', University of Göttingen, Germany, 1993.
[73] G. M. Sheldrick, 'SHELXL 97, Program for the Refinement of Crystal Structures', University of Göttingen, Germany, 1997.

Received October 5, 2000