Structures of Polypeptides from α-Amino Acids Disubstituted at the α-Carbon

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ABSTRACT: The structural preferences of homopeptides from Aib, Deg, Dpg, and Ac_nc (n = 3, 5, and 6) residues, as determined by conformational energy computations, X-ray diffraction analyses, and spectroscopic studies, are reviewed. The results obtained indicate that the 3_{10}-helix and the fully extended (C_5) conformation are preferentially adopted by long sequences of these $C^{\alpha,\alpha}$-disubstituted α-amino acid residues, depending upon bulkiness and nature (whether linear or cyclic) of their side chains.

I. Introduction

The confluence of the following motivational factors accounts for the numerous recent papers devoted to the investigation of the structural preferences of polypeptides from α-amino acids disubstituted at the α-carbon (Figure 1):

(i) The potential usefulness of these residues as a new type of conformational constraint in the synthesis of enzyme-resistant agonists and antagonists of bioactive peptides.[1-3]

(ii) The widespread presence of Aib (α-aminoisobutyric acid), the prototype of this family of residues, in the membrane-active, channel-forming quasi-ionophoric, naturally occurring peptaibol antibiotics.[4,5]

(iii) The high crystallinity of derivatives and peptides of these residues, thus allowing one to perform X-ray structural characterization of the N- and C-blocking groups, and C-activating groups (including elusive intermediates) commonly used in peptide synthesis.

(iv) The still open problem of the structures adopted by the corresponding homopolymers. This point is relevant to the exploitation of these compounds as precise molecular rulers or as protein scaffolding blocks in the de novo design of protein and enzyme mimetics.[6]

In this article the discussion will be focused mainly on the structures presented by well-characterized, monodispersed homopeptides from Aib, Deg ($C^{\alpha,\alpha}$-diethylglycine), Dpg ($C^{\alpha,\alpha}$-di-*n*-propylglycine), and Ac_nc (1-aminocycloalkane-1-carboxylic acid; n = 3, 5, and 6) of various main-chain lengths. Homopeptides from other amino acids of this family have not been investigated so far.

II. Poly(Aib)$_n$

A. Conformational Energy Computations. A number of conformational energy computations of Ac–Aib–NHMe (Ac, acetyl; NHMe, methylamino) showed that the presence of two methyl groups on the C^α carbon imposes a marked restriction on the available conformational space.[3,7-36] Folded (helical) structures of the 3_{10} (φ = ±60°, ψ = ±30°)[37] and α (φ = ±55°, ψ = ±45°) types are more stable than the C_7-conformation (φ = ±60°, ψ = ∓60°) and the fully extended (φ = ψ = ±180°) C_5-structure.[38] In addition, the energy difference between the 3_{10}- and α-helical structures is small. The nonplanar distortion of the peptide unit (ω angle) was also taken into account in the computations.[19,20] For the value $\Delta\omega$ = +6°,

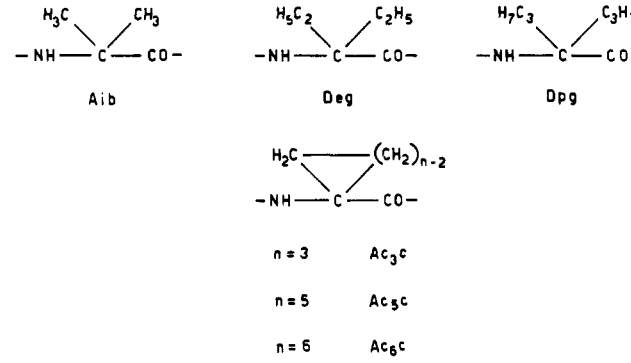

Figure 1. $C^{\alpha,\alpha}$-Disubstituted α-amino acids discussed in this work.

experimentally observed in the X-ray diffraction structures of oligopeptides containing the Aib–Aib sequence, a modified α-helix (φ = ±55°, ψ = ±60°), called the α′-helix, was found to be the most stable structure. In addition, it was demonstrated that the preferred helical conformation of Ac–(Aib)$_n$–NHMe is also sensitive to the molecular geometry (in particular to the bond angles involving the four substituents on the tetrahedral C^α-atom) assigned to this residue: a tetrahedral symmetric geometry for these substituents favors the α-helix, whereas an asymmetric geometry, derived from published X-ray diffraction structures, favors the 3_{10}-helix.[16]

The trend of the conformational energy recently computed for the extended and helical structures of Ac–Aib–NHMe as a function of the N–C^α–C′ bond angle (τ) indicates that in the whole range of values considered (which covers the complete set of X-ray values for peptides) the helical structure is more stable, although the energy difference decreases on decreasing the τ angle.[28,31-34] The most stable helical structure (τ = 111°, φ = ±53°, ψ = ±40°) is 0.3 kcal/mol lower in energy than the most stable extended conformation (τ = 107°, φ = ±173°, ψ = ±172°) but the small energy difference becomes compatible with the detection of a nonnegligible percentage of extended structures in Ac–Aib–NHMe. When the chain length is increased to Ac–(Aib)$_n$–NHMe ($n \geq 2$), a further stabilization is obtained only in the cases of helical structures, due to the formation of intramolecular H bonds between different residues, thus suggesting a shift of the conformational equilibrium toward such structures.

Computations performed for Ac–(Aib)$_n$–NHMe (n = 1–5 and 8) gave no evidence for the onset of α-helices, but

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they showed the preference for an early formation of incipient 3_{10}-helices.[30] On the other hand, by calculation of the energy of each type of helix for $(Aib)_n$ (n = 1–15), it has recently been demonstrated that the α-helix gains approximately 2 kcal/mol per residue in stability over the 3_{10}-helix.[27] Interestingly, in the early 1960s, Blout and Fasman[39] and Elliott et al.,[40] independently, from simple analysis of molecular models were able to show that unfavorable steric interactions are more significant in the α-helix of poly$(Aib)_n$ than in the 3_{10}-helix.

B. Solid-State Studies. All experimental investigations of poly$(Aib)_n$ have been carried out on polydispersed materials, prepared from *N*-carboxyanhydride polymerization.[41-46] X-ray diffraction powder photographs and unpolarized IR absorption results were originally interpreted in terms of an α-helix,[47,48] whereas more recent electron-diffraction photographs and polarized IR absorption data of oriented specimens produced evidence that the 3_{10}-helix is the preferred conformation.[49,50] A vibrational analysis of the normal modes of poly$(Aib)_n$ in both the α- and 3_{10}-helices showed significant differences only in the 300–900-cm^{-1} region: a comparison of observed with calculated frequencies supported the view of the occurrence of the 3_{10}-helical structure for the polypeptide.[51]

In order to obtain a clear-cut answer to the problem of the conformation adopted in the solid state by high molecular weight poly$(Aib)_n$, we performed an IR absorption, X-ray diffraction, and ^{13}C NMR investigation of the strictly monodispersed, fully protected $(Aib)_n$ peptides synthesized in our laboratory by solution methods to n = 12,[31] a value high enough to dramatically reduce the role of end-group effects that might be operative in low molecular weight oligomers.

The IR absorption spectra of the higher oligomers are similar and significantly simplified if compared with those of the lower oligomers.[31-33,52,53] From this study we were able to conclude that in the solid state the higher Aib oligomers adopt an ordered secondary structure characterized by strong H bonds. With regard to the type of secondary structure formed, the amide I band position ($\approx$1660 cm^{-1}) is *not* compatible with the onset of a β-structure (1630 cm^{-1}). It was not possible to discriminate between α- and 3_{10}-helices from the spectra in the amide A ($\approx$3350 cm^{-1}) and amide I regions, since the predicted frequencies are nearly the same. From an analysis of the amide V region (a broad band was observed near 680 cm^{-1}), where significant differences between the two types of helical structures are expected,[51] preliminary evidence has been obtained for the formation of the 3_{10}-helix at the n = 5–6 level.

To ascertain unambiguously the molecular structure of the $(Aib)_n$ homopeptides in the solid state, we took advantage of the high crystallinity of these compounds for an extensive X-ray diffraction investigation. In this connection, it is worth mentioning that the number of structures of Aib derivatives and homopeptides solved since 1978 is increasing extremely rapidly, much more than for any other amino acid residue, including proline (which also is conformationally restricted and gives easily crystallizable derivatives and peptides[54]). The complete list of X-ray structures, 37 out of 46 from our laboratories, is given in Table I.

The crystal structures of the N^{α}-blocked tripeptides show the formation of a folded structure stabilized by an 1 ← 4 intramolecular N–H···O=C H-bond characteristic of the type III (or the enantiomeric type III′) β-bend (or C_{10}-conformation).[38,89,90] This (incipient) 3_{10}-helix is fully developed at the pentamer level. Recently, we have been able to solve the structure of the terminally blocked $(Aib)_{10}$ oligomer.[85,86] This decapeptide adopts a perfect 3_{10}-helix, stabilized by eight consecutive intramolecular H bonds of the C_{10}-type (Figure 2). This is the first observation at atomic resolution of a regular polypeptide 3_{10}-helix as long as three complete turns. In this helical structure one peptide group is carried into the next to which it is directly chemically linked by a rotation of 112° and an axial translation of 1.96 Å. The pitch is 6.29 Å, and there are 3.21 residues/turn. The conformational angles are φ = ±54.1°, ψ = ±31.2°, and ω = ±176.1°. The mean value of the N···O distance of the eight intramolecular N–H···O=H bonds of the C_{10}-III (or C_{10}-III′) type is 3.04 Å, while those of the N–H···O and N···O=C angles are 159° and 130°, respectively. We believe that all these results, taken together, represent a decisive proof in favor of the 3_{10}-helix as the preferred conformation of poly$(Aib)_n$ in the solid state and that main-chain length is not an overriding factor in directing the type of helical folding in this homopeptide series.

In the course of the X-ray diffraction study of the $(Aib)_n$ homopeptides, a novel structure, typical of the C-terminal portion of a polypeptide chain, was detected.[74] The N-protected tripeptide Z–$(Aib)_3$–OH, in addition to one intramolecular H bond of the usual C_{10}-III (or C_{10}-III′) type involving the N(3)H group as the donor, exhibits an unusual intramolecular H bond of the O–H···O=C type with the OH group of the carboxylic acid moiety as the donor, giving rise to an *oxy analogue* of the C_{10}-III (or C_{10}-III′) type.[38] Interestingly, Z–$(Aib)_3$–OH monohydrate has the same overall conformation as that of the anhydrous

Table I
X-ray Diffraction Structures of Aib[a] Derivatives and Homopeptides

derivatives[b]	ref	homopeptides[b]	ref
1. Ac–Aib–OH	55	1. H_2^+–$(Aib)_2$–O^-	68
2. mClAc–Aib–OH	56	2. Z–$(Aib)_2$–OH	57, 60
3. dClAc–Aib–OH	56	3. Z–$(Aib)_2$–OH oxazolone	69
4. tClAc–Aib–OH	56	4. H–$(Aib)_2$–O*t*Bu	70
5. Tfa–Aib–OH	55	5. Tfa–$(Aib)_2$–O*t*Bu	70
6. Boc–Aib–OH	57–59	6. Boc–$(Aib)_2$–OBzl	71
7. Z–Aib–OH (α-form)	60	7. *c*–$(Aib)_2$	72
8. Z–Aib–OH (β-form)	60	8. H_2^+–$(Aib)_3$–O^-	68, 73
9. Fmoc–Aib–OH	61	9. Z–$(Aib)_3$–OH	74
10. Adoc–Aib–OH	62	10. Z–$(Aib)_3$–OH monohydrate	74
11. *p*BrBz–Aib–OH	63	11. Z–$(Aib)_3$–OH oxazolone	75
12. *p*BrBz–Aib–OH oxazolone	63	12. Ac–$(Aib)_3$–OMe	76
13. Ac–Aib–OMe	64	13. PhAc–$(Aib)_3$–OMe	65
14. *p*BrBz–Aib–OMe	63	14. Z–$(Aib)_3$–OMe	74
15. PhAc–Aib–OMe	65	15. Z–$(Aib)_3$–O*t*Bu	52, 57
16. Z–Aib–OBzl	64	16. Z–$(Aib)_4$–OH	52, 57
17. Z–Aib–OPcp	66	17. Tos–$(Aib)_4$–OMe	77
18. Boc–Aib–OPcp	66	18. Z–$(Aib)_5$–O*t*Bu	52, 57
19. $(Pht–Aib)_2O$	66	19. Tos–$(Aib)_5$–OMe	77–79
20. $(Z–Aib)_2O$	60	20. *p*BrBz–$(Aib)_6$–OMe	80
21. Ac–Aib–NHMe	67	21. Tos–$(Aib)_6$–OMe	77
		22. Z–$(Aib)_7$–OMe	81
		23. Z–$(Aib)_7$–O*t*Bu	82
		24. *p*BrBz–$(Aib)_8$–O*t*Bu	83, 84
		25. *p*BrBz–$(Aib)_{10}$–O*t*Bu	85, 86

[a] For the X-ray diffraction structure of H_2^+–Aib–O^- see refs 87 and 88. [b] The abbreviations used in this table are the following: Ac, acetyl; mClAc, monochloroacetyl; dClAc, dichloroacetyl; tClAc, trichloroacetyl; Tfa, trifluoroacetyl; PhAc, phenylacetyl; *p*BrBz, *p*-bromobenzoyl; Boc, (*tert*-butyloxy)carbonyl; Z, (benzyloxy)carbonyl; Fmoc, [(fluoren-9-ylmethyl)oxy]carbonyl; Adoc, (1-adamantyloxy)carbonyl; Pht, phthaloyl; Tos, tosyl; OMe, methoxy, OPcp, pentachlorophenyloxy; OBzl, benzyloxy; O*t*Bu, *tert*-butoxy; NHMe, methylamino.

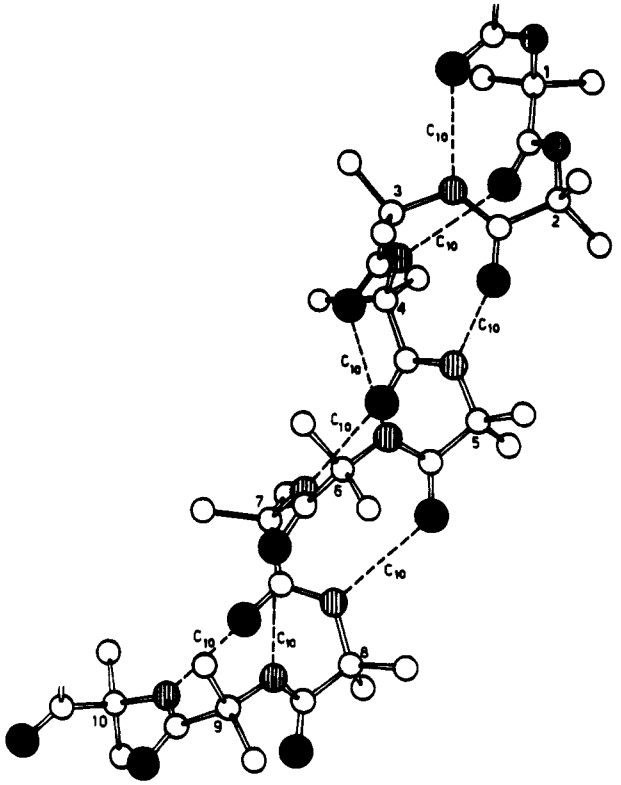

Figure 2. 3_{10}-Helix shown by *p*BrBz–(Aib)$_{10}$–O*t*Bu in the crystal state. The eight intramolecular H bonds of the C_{10}-type are indicated as dashed lines. Amino acid residues are numbered from the N-terminus. The *p*BrBz- and -O*t*Bu terminal groups have been omitted for clarity.

tripeptide. In the former, however, the water molecule is linked to the C-terminal OH group and to the C=O group of the Aib(1) residue, the same two groups involved in the formation of the oxy analogue of the C_{10}-III (or C_{10}-III′) type present in the latter, giving rise to a 12-membered, internal water bridge structure.[91]

Unusual conformations for an Aib residue, discovered in the X-ray structures listed in Table I, are the *semi*extended conformation ($\varphi = \pm 60°, \psi = \mp 140°$) [in Tfa–(Aib)$_2$–O*t*Bu[70] and in Boc–(Aib)$_2$–OBzl[71]] and the fully extended C_5-conformation [in H–(Aib)$_2$–O*t*Bu[70]]. It should be recalled that the only example available so far of a C_7-conformation for an Aib residue is the strained cyclotetrapeptide dihydrochlamydocin.[92]

In a recent high-resolution solid-state ^{13}C NMR analysis of the Z–(Aib)$_n$–OMe (n = 3–8) series, it has been reported that all the oligomers adopt an incipient or a fully developed 3_{10}-helical structure, as judged from the characteristic splitting of the C^β signals as well as the conformation-dependent displacements of the C^α and C′ peaks.[93]

C. Solution Studies. We have investigated the conformational preferences of the protected (Aib)$_n$ (n = 2–12) homopeptides in a solvent of low polarity ($CDCl_3$) at various concentrations and temperatures by using IR absorption (in the amide A and amide I regions) and ^1H NMR techniques.[31-34,52,53,84,86,94-97] Relevant conclusions are the following: (i) At 1.5 mM concentration intermolecular H bonding is negligible for all oligomers but the dodecamer, whereas self-association occurs at a concentration higher than 10 mM for the oligomers with $n \approx 3$. The NH groups involved in the H-bonding schemes as donors appear to be the urethane N(1)H and the peptide N(2)H groups, this phenomenon being particularly evident for the N(1)H. Further, association constants for the dimerization of selected oligomers were obtained from vapor-pressure osmometry measurements [as an example, the K_2 value for Z–(Aib)$_5$–O*t*Bu was found to be 11 m^{-1}]. (ii) In the absence of self-association, fully developed, thermally stable, intramolecularly H-bonded structures are formed at about the octamer level. The percentages of ordered secondary structure are still remarkably high for the oligomers from tetramer to heptamer, while clearly reduced, although still consistent, for the trimer.

Since all NH protons [beginning from the N(3)H proton] in the molecule form intramolecular H bonds, it may be concluded that the secondary structure adopted by the protected (Aib)$_n$ homopeptides in $CDCl_3$ is the 3_{10}-helix. A comparison between solid-state and solution ($CDCl_3$) results led us to conclude that the pleionomers (the monodispersed higher homologues having a main chain longer than the critical chain length for the onset of the ordered structure typical of the corresponding homopolymer[98]) from the conformationally restricted Aib residue form a 3_{10}-helical structure insensitive to environmental effects. Our results on the lowest Aib oligomers in $CDCl_3$ are in excellent agreement with those reported by other groups on short (Aib)$_n$ homopeptides differently blocked at the N- and C-termini.[99-104]

An instructive, although preliminary, conformational investigation of the Z–(Aib)$_n$–O*t*Bu (n = 2–12) series was carried out by thin-layer chromatography and melting point determinations.[105] This study revealed that the n = 5, 8, and 11 oligomers, where complete turns of 3_{10}-helices are expected, show higher R_F values than the other oligomers. In addition, their melting points are higher compared to those of oligomers having one Aib residue more or less. Thus, it appears that the most symmetrical structures are reflected in the analytical data.

For the first time we have been able to prove a $3_{10}^r \rightleftarrows 3_{10}^l$ helix enantiotopomerization [in the case of Z–(Aib)$_{10}$–O*t*Bu, via temperature-dependent ^{13}C NMR measurements between 200 and 300 K in dichloromethane].[106,107] At room temperature the conformationally uniform decapeptide shows a single Aib C^β signal. Upon cooling of the solution to 203 K two distinct signals originate out of the 20 prochiral C^β atoms of each enantiomeric 3_{10}-helix. The free energy of activation of the 3_{10}-helix interconversion was found to be 4.6 kJ mol^{-1} per Aib residue (at T_c = 265 K).

III. Poly(Deg)$_n$ and Poly(Dpg)$_n$

Unfavorable steric interactions between the NH group and the two β-substituents reduce the stability of the C_5-conformation of a $C^{\alpha,\alpha}$-disubstituted glycine. This effect, however, remains practically constant along the whole series Aib, Deg, and Dpg, as no significant interactions are connected to groups in the γ-position.[3,29,33,34,53,108-110] On the other hand, the stability of the helical structures decreases by lengthening the number of side-chain carbon atoms, since these chains are forced into strained conformations in order to reduce the interactions with the backbone. As a consequence, the energy difference between C_5 and helical structures decreases along the series. In contrast to Ac–Aib–NHMe, in Ac–Deg–NHMe and Ac–Dpg–NHMe the stability order of the (3_{10}- and α-) helices versus other peptide structures is strictly related to the τ angle; the fully extended (C_5) conformation, in particular, is favored for $\tau \leq 107°$. From this analysis it was possible to conclude that this type of conformation becomes more stable than the helical structures when both side-chain C^β atoms are substituted. The C_7 conformation is sensibly destabilized in all $C^{\alpha,\alpha}$-disubstituted α-amino acids by the presence of one of the two substituents in the unfavorable axial position relative to the H-bonded seven-membered ring.

Table II
X-ray Diffraction Structures of Deg, Dpg, and Ac_nc (n = 3, 5, and 6) Derivatives and Homopeptides[a]

derivatives[b]	ref	homopeptides[b]	ref
1. mClAc–Deg–OH	111	1. Tfa–$(Deg)_2$–O*t*Bu	110, 112
2. Tfa–Deg–OH	112	2. Tfa–$(Deg)_3$–O*t*Bu	110, 112
3. Ac–Deg–NHMe	113	3. Tfa–$(Deg)_4$–O*t*Bu	110, 112
4. mClAc–Dpg–OH	111	4. Tfa–$(Deg)_5$–O*t*Bu	110, 112
5. Tfa–Dpg–DBH	28, 109	5. Tfa–$(Dpg)_2$–DBH	28, 109
6. *p*BrBz–Ac_3c–OH	114	6. H–$(Ac_3c)_2$–OMe	114, 117
7. Piv–Ac_3c–OH	114	7. *c*–$(Ac_3c)_2$	114
8. Z–Ac_3c–OH	114	8. Ac–$(Ac_3c)_2$–OMe	117, 118
9. Boc–Ac_3c–OH (α-form)	114	9. Fmoc–$(Ac_3c)_2$–OMe	117, 118, 127
10. Boc–Ac_3c–OH (β-form)	114	10. *p*BrBz–$(Ac_3c)_3$=OMe	117, 118
11. *p*BrBz–Ac_3c–OH oxazolone	114	11. Boc–$(Ac_3c)_3$–OMe	117, 118
12. Z–Ac_3c–OMe	114	12. Boc–$(Ac_5c)_2$–NHMe	119
13. mClAc–Ac_5c–OH	115	13. *p*BrBz–$(Ac_5c)_4$–O*t*Bu	120, 121
14. *p*BrBz–Ac_5–OH	115	14. *p*BrBz–$(Ac_5c)_5$–O*t*Bu	120, 121
15. Fmoc–Ac_5c–OH	115	15. Z–$(Ac_5c)_6$–O*t*Bu	120, 121
16. Z–Ac_5c–OH	115	16. Z–$(Ac_6c)_2$–O*t*Bu	122
17. Z–Ac_5c–OH acetone solvate	115	17. Box–$(Ac_6c)_2$–OH	116
18. *p*BrBz–Ac_5c–OH oxazolone	115	18. Boc–$(Ac_6c)_3$–OMe	36
19. mClAc–Ac_6c–OH	116	19. Z–$(Ac_6c)_4$–O*t*Bu (α-form)	121, 123
20. Boc–Ac_6c–OH	116	20. Z–$(Ac_6c)_4$–O*t*Bu (β-form)	121, 123
21. $(Z–Ac_6c)_2O$	116	21. *p*BrBz–$(Ac_6c)_4$–O*t*Bu	121, 123
22. *p*BrBz–Ac_6c–OH oxazolone	116		

[a] For the X-ray diffraction structures of H_2^+–Ac_nc–O^- (n = 3, 5, and 6), see refs 114, 116, 124, 125, and 140. [b] The abbreviations used in this table are the following: Piv, pivaloyl; DBH, *N'*,*N'*-dibenzylacylhydrazino. All other abbreviations are the same as those listed in Table I.

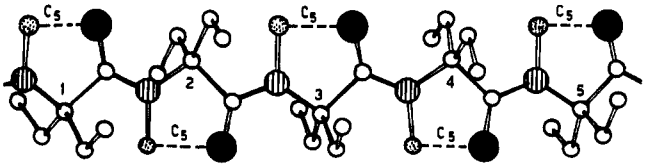

Figure 3. Fully extended conformation shown by Tfa–$(Deg)_5$–O*t*Bu in the crystal state. The five intramolecular H bonds of the C_5-type are indicated as dashed lines. Amino acid residues are numbered from the N-terminus. The Tfa- and -O*t*Bu terminal groups have been omitted for clarity.

In 19 out of 21 residues of Deg and Dpg derivatives and homopeptides (to the pentamer) examined to date by X-ray diffraction (Table II) the set of φ, ψ angles (values close to 180° and 180°) of each residue indicates the fully extended conformation (Figure 3). Interestingly, these peptides represent the first examples, apart from H_2^+–$(Gly)_3$–O^-,[126] in which consecutive C_5-conformations are experimentally observed. Further (although indirect) support for the existence of this type of *intra*molecular H-bonded conformation is given by the observation that the pertinent N–H and C=O groups are not involved in the *inter*molecular H-bonding schemes. This set of compounds allowed us to characterize the C_5-conformation in detail for the first time. In particular, the mean N···O and H···O intramolecular distances are 2.54 and 2.00 Å, respectively. The τ angle is narrowed to 102.8°. The ethyl and *n*-propyl side chains of Deg and Dpg residues, respectively, are also fully extended, in order to relieve the unfavorable intramolecular main-chain to side-chain and side-chain to side-chain interactions. For the first time, in Tfa–$(Deg)_3$–O*t*Bu, a crystallographic planar peptide skeleton for the main-chain atoms ($\varphi = \psi = \omega$ 180°) has been observed; in fact, the molecules lie on the mirror plane and the two side chains of each residue are each the mirror image of the other.[110,112]

IR absorption and ^1H NMR investigations of the Deg and Dpg derivatives and peptides in $CDCl_3$ solution strongly support the view that under these experimental conditions the largely prevailing conformation is again the fully extended conformation.[96,108-111] In particular, intramolecular H bonding was found to be the dominant factor and the significant spectroscopic parameters were shown to be concentration independent and to exhibit remarkably small variations as the main chain is elongated, the temperature is raised, and perturbing agents are added. The restricted mobility of the Deg and Dpg side chains of all homopeptides is demonstrated by the magnetic nonequivalence of the β-methylene protons in the ^1H NMR spectra.

IV. Poly(Ac_nc)$_n$

The great versatility of the family of $C^{\alpha,\alpha}$-disubstituted glycines has been demonstrated by conformational energy computations of Ac–$(Ac_3c)_n$–NHMe (n = 1–3)[127-130] and X-ray diffraction analyses of a variety of peptides of this residue to the tetramer level[114,117,118,127] (Table II). These studies clearly indicate the marked structural preferences of the three-membered ring Ac_3c for the "bridge" region of the conformational space, in particular for $\varphi,\psi \approx \pm 90°$, 0°, that is, for position i + 2 of type-I (I') and type-II (II') β-bends (however, no experimental evidence has been found so far for Ac_3c in the latter type of bend). This residue can also be accommodated in *distorted* type-III (III') β-bends and 3_{10}-helices ($\varphi,\psi \approx \pm 60°, \pm 30°$). The most striking feature of the Ac_3c φ,ψ map is the absence of the fully extended conformation. This phenomenon is essentially due to steric repulsions between the carbonyl moiety and the β-methylene hydrogens.

As for the average (asymmetric) geometry of Ac_3c, the carbon atoms of the cyclopropyl ring form an isosceles triangle, with the *distal* bond opposite to the substituents being approximately 0.02 Å shorter than the *vicinal* equal sides. As expected for a three-membered ring, the *endo*cyclic bond angles have values close to 60°, but the angle at C^α is slightly smaller than the angles at $C^{\beta L}$ and $C^{\beta D}$. Concomitantly, the mean value for the *exo*cyclic angle (116.7°) is significantly larger than the regular tetrahedral angle, an indication of the spread between the amino and carbonyl functions on the cyclopropyl ring. The N–C^α (1.43 Å) and C^α–C' (1.49 Å) bond lengths are significantly shortened compared to C^α-*mono*substituted α-amino acids and the Aib residue,[64] a clear indication of the conjugative ability of the cyclopropyl moiety.

Interestingly, the preferred conformations (regular type-III β-bends and 3_{10}-helices) and the typical values for the τ angle (≈111°) theoretically and experimentally (X-ray diffraction) found for Ac_5c and Ac_6c residues with cyclic moieties significantly larger than that of Ac_3c, closely parallel those of the prototype Aib.[36,115,116,119-121,123] In the derivatives and homopeptides (to the hexamer) examined (Table II) the cyclopentyl rings adopt either an envelope or half-chair conformation, while the cyclohexyl rings are almost perfect chair conformations with predominantly axial arrangement of the amino substituent. The effect of the side-chain conformational changes produced by cyclization upon preferred backbone structure is evident from a comparison of the results for the Ac_5c peptides with those for the open-chain carbon atoms. Again, the same preferred conformations (β-bends and 3_{10}-helices) as those found in the crystal state, were detected in $CDCl_3$ solution.[36,119,121,127,128,131,132] Polymeric $(Ac_6c)_n$ was prepared a long time ago either by *N*-car-

boxyanhydride[133] or by the dipeptide oxazolone[134] method, but its structure has never been investigated in detail.

V. Conclusion

The results of a number of theoretical and experimental studies indicate that the 3_{10}-helix and the fully extended C_5-conformation are preferentially adopted by oligomers of the Aib, Deg, Dpg, and Ac_nc (n = 3, 5, and 6) residues as a function of the number of carbon atoms and the nature (whether linear or cyclic) of their side chains. It is evident that the structural versatility of homopeptides from $C^{\alpha,\alpha}$-disubstituted glycines is now just beginning to emerge. Additional studies are needed to complete the picture. In particular, the well-established sensitivity of conformation to geometry (τ bond angle) requires a thorough analysis of peptides containing Ac_4c and Ac_9c residues: the former (four-membered ring containing α-amino acid) is expected to exhibit values for the τ angle significantly larger than the regular tetrahedral value,[135] while the latter is expected to show a markedly smaller value because of the expansion of the *endo*cyclic C–C–C angles of the nine-membered ring to relieve the transannular compression of the H atoms.[136,137] An α-helical structure incorporating cyclic $C^{\alpha,\alpha}$-dialkylated glycines (in particular Ac_5c and Ac_6c residues) still remains to be reported.

The fully extended conformation should be examined more extensively by studying peptides from other $C^{\alpha,\alpha}$-disubstituted glycines with linear, symmetrical side chains, e.g., di-*n*-butylglycines.[138]

The type of preferred helical structure and the relationship between absolute configuration at the C^{α}-atom and helix-screw sense should be investigated theoretically and experimentally by examining *iso*- and *syndio*tactic homopeptides from various *chiral* residues of this family.[76,139] Studies along these lines are currently in progress in our laboratories.

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