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Stabilization of two smallest possible diastereomeric β-hairpins in a water soluble tetrapeptide containing non-coded α-amino isobutyric acid (Aib) and *m*-amino benzoic acid

Anita Dutt [a], Arpita Dutta [a], Sudeshna Kar [a], Pradyot Koley [a], Michael G.B. Drew [b], Animesh Pramanik [a,*]

[a] Department of Chemistry, University of Calcutta, 92, A.P.C. Road, Kolkata 700 009, India
[b] School of Chemistry, The University of Reading, Whiteknights, Reading RG6 6AD, UK

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ABSTRACT

Single crystal X-ray diffraction study reveals that the water soluble tetrapeptide H_2N–Ile–Aib–Leu–*m*-ABA–CO_2H, containing non-coded Aib (α-amino isobutyric acid) and *m*-ABA (*meta*-amino benzoic acid), crystallizes with two smallest possible diastereomeric β-hairpin molecules in the asymmetric unit. Although in both of the molecules the chiralities at Ile(1) and Leu(3) are S, a conformational reversal in the back bone chain is observed to produce the β-hairpins with β-turn conformations of type II and II′. Interestingly Aib which is known to adopt helical conformation, adopts unusual semi-extended conformation with ϕ: −49.5(5)°, ψ: 135.2(5)° in type II and ϕ: 50.6(6)°, ψ: −137.0(4)° in type II′ for occupying the $i+1$ position of the β-turns. The two hairpin molecules are further interlocked through intermolecular hydrogen bonds and electrostatic interactions between $-CO_2^-$ and $-^+NH_3$ groups to form dimeric supramolecular β-hairpin aggregate in the crystal state. The CD measurement and 2D NMR study of the peptide in aqueous medium support the existence of β-hairpin structure in water.

1. Introduction

β-Hairpins are known to participate in many important biological recognition processes such as protein–protein [1], protein–RNA [2,3], and protein–DNA interactions [4,5]. Alzheimer's disease [6–8], prion diseases [9–11], the IgE-mediated allergic response [12], the interaction of bacterial cell surface-associated protein with IgG [13–15], and HIV gp 120 binding to human T-cell surface protein CD4 [16,17] are some examples involving the β-hairpin secondary structure element in biological processes. In spite of its significance, the principles underlying hairpin stability are still unresolved, perhaps because of the general difficulty of investigating hairpins, due to their low solubility and high propensity to aggregate [18–23]. Despite these obstacles, several examples of naturally occurring β-hairpins and hairpin mimetics have been reported [24–28].

Various factors such as the type of β-turn [16,20,23], the β-sheet-forming tendencies of amino acids [29–31], the length of the peptide strands [32–34], steric factors [35], the role of interchain hydrogen bonds [36–38] and electrostatic interactions control the hairpin stability [20,39]. But the relative importance of these factors is not fully understood as yet. In most β-hairpin designs, the strand regions contain hydrophobic residues, while the D-Pro-Gly segment, which in principle can adopt both II′ and I′ turns, is most often used to form the tight turn [40–43]. To a lesser extent segments like Asn-Gly and Aib-D-Ala have also been used to facilitate the formation of I′ turn in β-hairpin designs [44–47]. Solution phase studies by Kunwar et al. show that the β-hairpins of terminally protected octapeptides tolerate insertion of *m*-amino benzoic acid and also permits accomodation of both enantiomers of Pro-Gly turn motifs [48,49]. Although there are several examples of large β-hairpin designs in the literature [40–49] stabilization of β-hairpin in tetrapeptide is rare. Designing the smallest possible β-hairpin by inserting *m*-amino benzoic acid, a substituted γ-amino butyric acid with an all *trans* extended configuration and also without using D-Pro-Gly segment or any rigid turn mimetic molecules will provide more insights about the various cooperative weak interactions responsible for β-hairpin nucleation and stabilization. Therefore our attempt was to design and synthesize a terminally deprotected water-soluble tetrapeptide H_2N–Ile(1)–Aib(2)–Leu(3)–*m*-ABA(4)–CO_2H(**I**) (Aib, α-amino isobutyric acid; *m*-ABA, *meta*-amino benzoic acid) to examine the hairpin formation both in the crystal state and in water (Fig. 1). The centrally placed Aib(2)–Leu(3) fragment, where the achiral Aib is a overwhelmingly constrained residue to adopt helical conformation [50–57], is expected to produce the turn required for a β-hairpin nucleation.

* Corresponding author. Tel.: +91 33 2484 1647; fax: +91 33 2351 9755.
E-mail address: animesh_in2001@yahoo.co.in (A. Pramanik).

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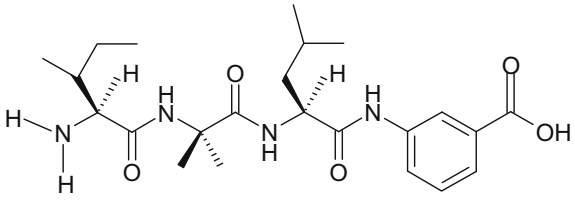

Fig. 1. Schematic representation of peptide **I**.

2. Experimental

2.1. Synthesis of peptides

The protected tetrapeptide was synthesized by conventional solution phase methods using racemization free fragment condensation strategy [58]. The Boc group was used for N-terminal protection and the C-terminus was protected as methyl ester. Couplings were mediated by *N-N′*-dicyclohexylcarbodiimide/1-hydroxybenzotriazole (DCC/HOBt) [58]. Deprotection of methyl ester was carried out using saponification method and the Boc group was deprotected by TFA (trifluoroacetic acid). The final product was purified by column chromatograghy using silicagel (100–200 mesh) as stationary phase and chloroform methanol mixture as the eluent. All the intermediates and purified final compound have been fully characterised by various spectroscopic techniques like NMR, IR and mass spectrometry. Single crystal suitable for X-ray diffraction study of peptide **I** was obtained from water–ethanol solution by slow evaporation.

2.1.1. Boc–Ile–Aib–Leu–m-ABA–OMe (*1*)

Boc–Ile–Aib–Leu–OH [52] (0.96 g, 2.23 mmol) was dissolved in DMF (3 ml). *m*-ABA–OMe (0.61 g, 4.46 mmol) obtained from its hydrochloride was added followed by DCC (0.69 g, 3.35 mmol) and HOBt (0.36 g, 2.68 mmol). The reaction mixture was stirred at room temperature for 5 days. The precipitated *N-N′*-dicyclohexylurea (DCU) was filtered and diluted with ethyl acetate. The organic layer was washed with excess of water, 1 M HCl (3×30 ml), 1 M Na_2CO_3 solution (3×30 ml) and again with water. The solvent was then dried over anhydrous Na_2SO_4 and evaporated in *vacuo*, giving a light yellow solid. Purification was done using silica gel as stationary phase and ethyl acetate–petroleum ether mixture as the eluent.

Yield: 1.11 g (94.0%). Mp = 110–111 °C; IR (KBr): 3306, 1723, 1665, 1528 cm^{-1}; ^1H NMR 300 MHz ($CDCl_3$, δ ppm): 9.17 (*m*-ABA(4) NH, 1H, s); 8.52 (*m*-ABA(4) Ha, 1H, s); 8.15 (*m*-ABA(4) Hd, 1H, d, $J = 7.9$ Hz); 7.71 (*m*-ABA(4) Hb, 1H, d, $J = 7.7$ Hz); 7.33 (*m*-ABA(4) Hc, 1H, t, $J = 7.9$ Hz); 7.24 (Leu(3) NH, 1H, d, $J = 7.8$ Hz); 6.70 (Aib(2) NH, 1H, s); 5.21 (Ile(1) NH, 1H, d); 4.54 ($C^{\alpha}H$ of Leu(3), 1H, m); 3.86 (–OCH_3, 3H, s); 3.80 ($C^{\alpha}H$ of Ile(1), 1H, m); 1.84–1.89 ($C^{\beta}Hs$ of Ile(1) and Leu(3) 2H, m); 1.59 ($C^{\beta}Hs$ of Aib(2), 6H, s); 1.49 (Boc–CH_3s, 9H, s); 1.23–1.30 ($C^{\gamma}Hs$ of Ile(1), 3H, m); 0.92–0.93 ($C^{\gamma}Hs$ of Ile(1) and $C^{\delta}Hs$ of Ile(1) & Leu(3), 9H, m); ^{13}C NMR 75 MHz ($CDCl_3$, δ ppm): 173.80, 171.84, 171.30, 167.09, 156.95, 139.09, 130.41, 128.53, 124.75, 124.52, 121.15, 81.28, 60.96, 57.02, 52.82, 51.92, 40.04, 36.49, 28.10, 27.40, 25.52, 25.11, 23.78, 23.25, 22.57, 20.76, 15.63, 11.51; Anal. Calcd. for $C_{29}H_{46}N_4O_7$ (562.69): C, 61.89; H, 8.24; N, 9.95%. Found: C, 61.75; H, 8.08; N, 9.80%.

2.1.2. Boc–Ile–Aib–Leu–m-ABA–OH (*2*)

1 (1.0 g, 1.77 mmol) was dissolved in methanol (15 ml) and 2 M NaOH (8 ml) was added. The reaction mixture was stirred at room temperature for 2 days. The progress of the reaction was monitered by TLC. After completion of the reaction the methanol was evaporated. The residue obtained was diluted with water and washed with diethyl ether. The aqueous layer was cooled in ice and neutralised by 2 M HCl and extracted with ethyl acetate. The solvent was evaporated in vacuo to give a white solid.

Yield: 0.87 g (90.0%). Anal. Calcd. for $C_{28}H_{44}N_4O_7$ (548.664): C, 61.29; H, 8.08; N, 10.21%. Found: C, 61.42; H, 8.25; N, 10.38%.

2.1.3. H_2N-Ile(1)-Aib(2)-Leu(3)-m-ABA(4)-CO_2H(Peptide *I*)

To 0.6 g (1.09 mmol) of **2,** 2 ml of TFA was added at 0 °C. The reaction mixture was then stirred at room temperature and the progress of deprotection of Boc-group was determined by thin layer chromatography (TLC). After 6 h TFA was removed completely under vacuo and the residue was taken in water, washed with diethyl ether. The aqueous part was then dried under vacuo to yield **I** as a white solid**.** Purification was done using silica-gel as the stationary phase and chloroform–methanol mixture as the eluent. Single crystals of the compound were grown from water–ethanol mixture by slow evaporation.

Yield: 0.49 g (90.0%). Mp = 204–206 °C; IR (KBr): 3333,1666,1664 cm^{-1}; ^1H NMR 300 MHz (d_6-DMSO, δ ppm): 9.77 (*m*-ABA(4) NH, 1H, s); 8.55 (Leu(3) NH,1H, bs); 8.41 (*m*-ABA(4) Ha, 1H, s), 8.00 (Aib(2) NH, 1H, s), 7.94(*m*-ABA(4) Hd, 1H, d, $J = 7.5$ Hz), 7.60 (*m*-ABA(4) Hb, 1H, d, $J = 7.2$ Hz), 7.32 (*m*-ABA(4) Hc, 1H, t, $J = 7.8$ Hz), 4.33–4.35 ($C^{\alpha}Hs$ of Ile(1) & Leu(3), 2H, m), 1.55–1.80 ($C^{\beta}Hs$ of Ile(1) and Leu(3) 2H, m), 1.37–1.46 ($C^{\beta}Hs$ of Aib(2), 6H, s), 1.03–1.30 ($C^{\gamma}Hs$ of Ile(1) and Leu(3), 3H, m), 0.81–0.85 ($C^{\gamma}Hs$ of Ile(1) and $C^{\delta}Hs$ of Ile(1) & Leu(3), 12H, m); Anal. Calcd. for $C_{23}H_{36}N_4O_5$ (448.54): C, 61.58; H, 8.09; N, 12.49%. Found: C, 61.42; H, 7.92; N, 12.33%. HR–MS (M^+Na^+) = 471.29, Mcalcd $(M^+Na)^+$ = 471.55.

2.2. FT-IR spectroscopy

IR spectra were examined using a Perkin Elmer – 782 model spectrophotometer. The solid-state FT-IR measurements were performed using the KBr disk technique.

2.3. NMR experiments

The ^1H NMR study was recorded on a Bruker Avance 500 model spectrometer operating at 300 MHz, respectively. The 2D experiment was carried out in D_2O on a Bruker DRX 500 MHz equipped with a 5 mm broadband inverse probe head. The peptide concentration was in the range 5–10 mM in D_2O for ^1H NMR and 20–30 mM for 2D NMR measurements.

2.4. Circular dichroism spectroscopy

Aqueous solution of peptide **I** (1.5 mM) was used for obtaining the CD spectrum. Far-UV CD measurements were recorded at 25 °C with a 0.5 s averaging time, a scan speed of 50 nm/min, using a JASCO spectropolarimeter (*J* 720 model) equipped with a 0.1 cm pathlength cuvette. The measurements were taken at 0.2 nm wavelength intervals, 2.0 nm spectral bandwidth and five sequential scans were recorded for the sample.

2.5. Mass spectrometry

Mass spectra of peptide **I** was recorded on HEWLETT PACKARD Series 1100MSD and Micromass Qtof Micro YA263 mass spectrometers by positive mode electro spray ionization.

2.6. Crystal data for peptide *I*

$C_{46}H_{90}N_8O_{14.5}$, $M = 987.19$, monoclinic, spacegroup C2, $Z = 4$, $a = 30.820(3)$ Å, $b = 8.603(1)$ Å, $c = 20.994(5)$ Å, $\beta = 109.41(2)°$,

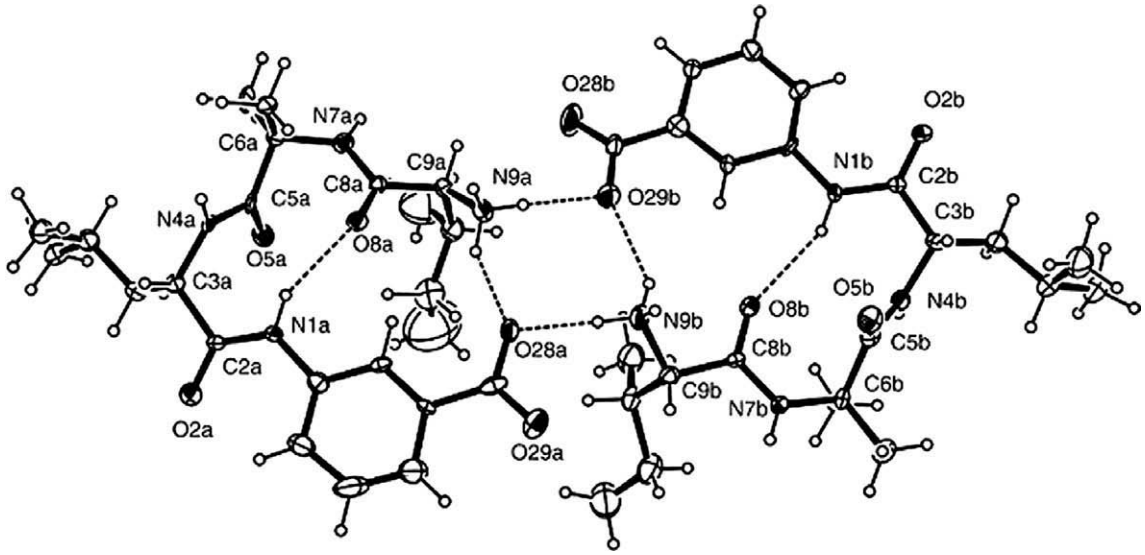

Fig. 2. The hairpin structure of peptide **I** with ellipsoids at 25% probability. Two hairpin molecules **A** and **B** are interlocked to form dimeric supramolecular β-hairpin aggregate. Hydrogen bonds are shown as dotted lines. For clarity solvent molecules are omitted.

U = 5249.9(14) Å^3, d_{calcd} = 1.238 g cm^{-3}. The crystal was positioned at 50 mm from the CCD. Three hundred and twenty-one frames were measured with a counting time of 20 s. Data analysis was carried out with the Crysalis program [59]. The structure was solved using direct methods with the Shelxs97 program [60–63]. The non-hydrogen atoms were refined with anisotropic thermal parameters. The hydrogen atoms bonded to carbon and nitrogen were included in geometric positions and given thermal parameters equivalent to 1.2 times (1.5 times for methyl groups) those of the atom to which they were attached. The structure was refined on F^2 using Shelxl97 to R1 0.0771, wR2 0.1367 for 4535 reflections with $I > 2\sigma(I)$. The data have been deposited at Cambridge Crystallographic Data Center with reference number CCDC 674779.

3. Results and discussion

3.1. Peptide conformations in the crystal state

The crystal structure of peptide **I** contains two molecules (**A** and **B**) in the asymmetric unit with different conformations. In both the molecules the chiralities at Ile(1) and Leu(3) are S. The molecules **A** and **B** are joined together by two intermolecular hydrogen bonds to form a molecular dimer (Fig. 2). Molecule **A** adopts a folded conformation corresponding to a slightly distorted type II β-turn structure with Aib(2) and Leu(3) occupying the $i + 1$ and $i + 2$ positions, respectively. The torsion angles at Aib(2) and Leu(3) were found to be ϕ_2: −49.5(5)°, ψ_2: 135.2(5)° and ϕ_3: 69.1(5)°, ψ_3: 14.4(5)°, respectively (Table 1), which deviate significantly from the ideal values for a type II β-turn ϕ_1: −60°, ψ_1: 120° and ϕ_2: 80°, ψ_2: 0°. As a result a weak intramolecular hydrogen bond between Ile(1)–CO and *m*-ABA(4)–NH (N1A–H1A···O8A, 2.20 Å, Table 2) is observed. Interestingly another intramolecular hydrogen bond between Ile(1)–$^+NH_3$ and *m*-ABA(4)–CO_2^- (N9A–H9A2···O28A, 2.02 Å, Table 2) is built up to produce the smallest possible β-hairpin structure. The planar and extended geometry of *m*-amino benzoic acid helps to form interstrand hydrogen bonds. The ψ_1 value at Ile(1) is found to be 151.1(4)°, which corresponds to an extended structure.

The second molecule in the asymmetric unit **B** adopts a type II′ β-turn structure, diastereomeric to **A**. The torsion angles at Aib(2) and Leu(3) were found to be ϕ_2: 50.6(6), ψ_2: −137.0(4) and ϕ_3:−97.4(4), ψ_3: 14.7(2), respectively (Table 1). This molecule **B** also contains the same two intramolecular hydrogen bonds that were observed in molecule **A** as listed in Table 2 and illustrated in Fig. 2. Planar *m*-amino benzoic acid modulates the loop conformation to a type II′ turn which is more flattened in nature compared to the twisted I′ counterpart [47]. As a result Aib which is known to adopt helical conformation [50–57], adopts unusual semi-extended conformation with ϕ_2: −49.5(5)°, ψ_2:135.2(5)° in type II and ϕ_2: 50.6(6)°, ψ_2: −137.0(4)° in type II′ **β-**turns. Although in both of the molecules **A** and **B** the chiralities at Ile(1) and Leu(3)

Table 1
Selected torsion angles (°) for peptide **I**.

	Molecule A	Molecule B
N9–C9–C8–N7(ψ_1)	151.1(4)	145.2(4)
C9–C8–N7–C6(ω_1)	176.5(4)	168.0(4)
C8–N7–C6–C5(ϕ_2)	−49.5(5)	50.6(19)
N7–C6–C5–N4(ψ_2)	135.2(5)	−137.0(4)
C6–C5–N4–C3(ω_2)	174.3(4)	−172.8(4)
C5–N4–C3–C2(ϕ_3)	69.1(5)	−97.4(4)
N4–C3–C2–N1(ψ_3)	14.4(5)	14.7(2)
C3–C2–N1–C21(ω_3)	174.2(4)	175.5(5)
C2–N1–C21–C22	169.2(4)	−161.3(4)
C22–C23–C27–O28	−14.7(7)	9.9(8)

Table 2
Selected intra and intermolecular hydrogen bonding parameters for **I**.

D–H···A	H···A/Å	D···A/Å	D–H···A/°
N9A–H9A1···O29B	1.85	2.724	170
N9A–H9A2···O28A	2.02	2.792	143
N9B–H9B1···O29B	2.20	2.946	142
N9B–H9B3···O28A	1.86	2.740	179
N1B–H1B···O8B	2.30	3.086	153
N1A–H1A···O8A	2.20	3.041	149
N9A–H9A3···O1(w)[a]	1.96	2.831	165
N7A–H7A···O2(w)[b]	2.15	2.893	144
N4A–H4A···O2A[c]	2.12	2.874	146
N9B–H9B2···O5(w)[d]	2.24	3.041	149
N4B–H4B···O2B[e]	2.20	3.008	156
N7B–H7B···O29A[f]	2.00	2.854	175

Symmetry elements: [a] $x, y + 1, z$; [b] $-x + 1, y + 1, -z + 1$; [c] $-x + 1/2, y + 1/2, -z + 1$; [d] $x, y + 1, z$; [e] $-x + 3/2, y - 1/2, -z + 2$; [f] $-x + 1, y, -z + 2$.

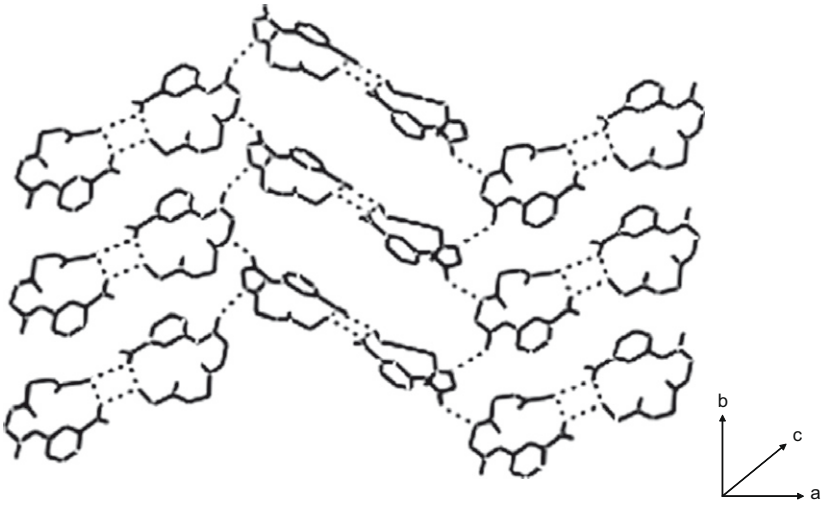

Fig. 3. Packing arrangement of peptide **I** showing the formation of supramolecular β-hairpin structure. Side chains of amino acids and solvent water molecules are omitted for clarity. Hydrogen bonds are shown as dotted line.

are S, a conformational reversal in the main chain is observed to produce the β-hairpins with β-turn conformations of type II and II′. Generation of two diastereomeric type II′ and II turns by the same hairpin sequence has not been observed previously. The present result also reveals that interstrand electrostatic interactions are important for stabilization of small β-hairpin structure.

In the asymmetric unit the two β-hairpin molecules **A** and **B** are interlocked through intermolecular hydrogen bonds (N9A–H9A1···O29B, N9B–H9B3···O28A, Table 2) and electrostatic interactions between $-CO_2^-$ and $-^+NH_3$ groups to form a dimeric supramolecular β-hairpin structure (Fig. 2). It is not clear whether the individual hairpins are stable enough to exist separately or that the stabilization provided by the dimeric assembly is necessary. In the crystal the dimers are further hydrogen bonded (N4A–H4A···O2A, N4B–H4B···O2B and N7B–H7B···O29A) to form a corrugated β-sheet like structure (Fig. 3, Table 2). The crystal structure contains several solvent water molecules. These form a few intermolecular hydrogen bonds (Table 2), and facilitate the formation of the supramolecular β-sheet structure.

3.2. Solution phase conformations

Sequence specific assignments of peptide **I** were readily achieved using a combination of DQF COSY and NOESY methods in D_2O [63]. The appearance of most of the NH resonances of peptide **I** at extremely low field (<8.0 ppm) is indicative of a hairpin conformation in which the backbone protons are expected to be deshielded [64].

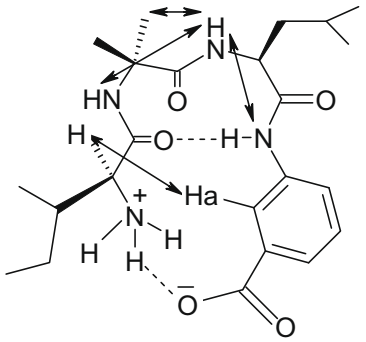

Fig. 4. Schematic representation of the proposed β-hairpin structure in peptide **I**. The hydrogen bonds are shown as dotted lines. Observed NOEs are highlighted by double edged arrows in water (500 MHz).

Further attempts to characterize the conformation in D_2O reveal that peptide **I** exhibits some important NOEs, namely Aib(2)NH ↔ Aib(2)C^{β}Hs, Leu(3)NH ↔ Aib(2)C^{β}Hs and Ile(1)C^{α}H ↔ *m*-ABA(4) Ha indicative of a β-hairpin conformation nucleated by a β-turn with Aib–Leu as the corner residues (Figs. 4 and 5). Moreover the existence of the interstrand NOE between the alpha protons of isoleucine and *m*-ABA Ha proton (Ile(1)C^{α}H ↔ *m*-ABA(4) Ha) is diagnostic of a hairpin conformation in which the strand regions are occupied by isoleucine and *m*-amino benzoic acid having high β-sheet propensity (Fig. 5) [65].

The conformation of peptide **I** was probed further in solution phase by far-UV CD measurement in water. The CD pattern of peptide **I** is presented in Fig. 6. Notably the spectrum of the water soluble peptide reveals a broad band at 210–225 nm with a distinct negative maximum at 217 nm and a cross-over point at nearly 205 nm, differing slightly from that observed for conventional β-hairpin peptides which shows a negative maximum at 220 nm. Indeed such anamolous CD patterns are observed for *m*-ABA containing peptides [48–49]. Thus such anamolous behaviour may be attributed due to the presence of *m*-ABA which can contribute to the CD with its aromatic chromophore. The self aggregation of haipins in water may also cause some changes in the CD pattern. Nevertheless, in addition to the NMR experiment, the CD data too strongly favor the conclusion that peptide **I** adopts a hairpin conformation in solution. Due to anamolous behaviour of the CD spectra of peptide **I** the co-existence of both the hairpin isomers **A** and **B** cannot be established in the solution phase unequivocally.

4. Conclusions

The present study establishes that it is possible to stabilize a smallest possible β-hairpin structure in a water soluble tetrapeptide containing non-coded α-amino isobutyric acid (Aib) and *meta*-amino benzoic acid. The result indicates that Aib–Leu segment can be utilized to generate turns for β-hairpins. The all *trans* extended configuration of *m*-amino benzoic acid generates flat non-twisted strand in the hairpin which is responsible for modulating the turn conformations in the hairpin. As a consequence Aib which is known to adopt helical conformation, adopts unusual semi-extended conformation with ϕ: −49.5(5)°, ψ: 135.2(5)° in type II and ϕ: 50.6(6)°, ψ: −137.0(4)° in type II′ **β**-turns of the

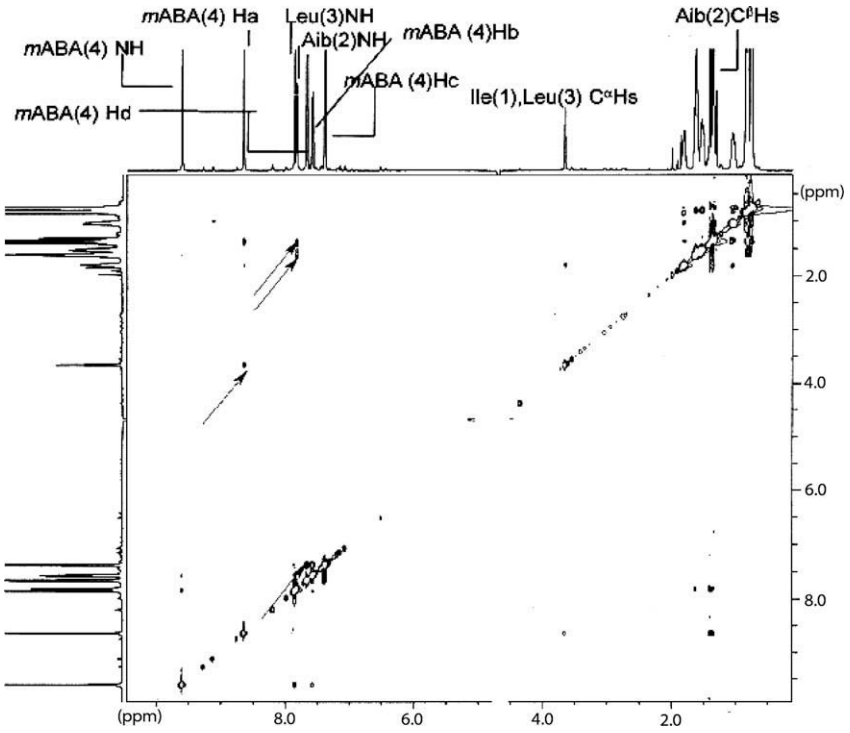

Fig. 5. NOESY (500 MHz) spectrum of peptide **I** in D_2O (peptide concentration: 1×10^{-2} M) exhibiting the cross peaks diagnostic of small hairpin conformation.

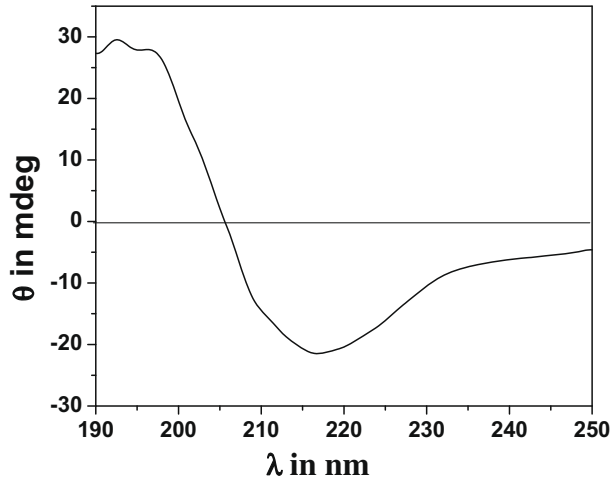

Fig. 6. CD spectra of peptide **I** in water, peptide concentration: 1.5 mM.

hairpins. The study also reveals that interstrand electrostatic interactions are necessary for stabilization of small β-hairpins with minimum number of amino acid residues. In the present study the formation of dimeric supramolecular hairpins aggregate through the self-assembly of small β-hairpins is a unique structural display.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.molstruc.2009.03.027.

Supplementary data contains a capped stick model of hairpin structure, ^{1}H NMR spectrum, Mass spectrum, an enlarged NOESY spectrum and crystallographic data in CIF format.

References

[1] N.J.C. Strynadka, S.E. Jensen, P.M. Alzari, M.N.G. James, Nat. Struct. Biol. 3 (1996) 290.
[2] J.D. Puglishi, L. Chen, S. Blanchard, A.D. Frankel, Science 270 (1995) 1200.
[3] G. Variani, Acc. Chem. Res. 30 (1997) 189.
[4] P.E. Floreancing, S.E. Swalley, J.W. Trauger, P.B. Dervan, J. Am. Chem. Soc. 115 (1993) 5887.
[5] R.E. Bremer, J.W. Szewczyk, E.E. Baird, P.B. Dervan, Bioorg. Med. Chem. 8 (2000) 1947.
[6] L.C. Serpell, C.C.F. Blake, P.E. Fraser, Biochemistry 39 (2000) 13269.
[7] S. Ohnisiki, A. Koide, S. Koide, J. Mol. Biol. 301 (2000) 477.
[8] D.R. Booth, M. Sunde, V. Bellotti, C.V. Robinson, W.L. Hutchinson, P.E. Fraser, P.N. Hawkins, C.M. Dodson, S.E. Radford, C.C.F. Blake, B.M. Pepys, Nature 385 (1997) 787.
[9] J. Collinge, Annu. Rev. Neurosci. 24 (2001) 519.
[10] K.M. Pan, M. Baldwin, J. Nguyen, M. Gasset, A. Serban, D. Groth, I. Mehlhorn, Z. Huang, R.J. Fletterick, F.E. Cohen, S.B. Prusiner, Proc. Natl. Acad. Sci. USA 90 (1993) 10962.
[11] R. Reik, S. Hornemann, G. Wider, M. Billeter, R. Glockshuber, K. Wüthrich, Nature 382 (1996) 180.
[12] J.M. McDonnell, D. Fushman, S.M. Cahill, B.J. Sutton, D. Cowburn, J. Am. Chem. Soc. 119 (1997) 5321.
[13] J.P. Derrick, D.B. Wigley, Nature 359 (1992) 752.
[14] C. Boutillon, R. Wintjens, G. Lippens, H. Drobecq, A. Tartar, Eur. J. Biochem. 231 (1995) 166.
[15] E.L. McCallister, E. Alm, D. Baker, Nat. Struct. Biol. 7 (2000) 669.
[16] A. Cochran, R.T. Tong, M.A. Starovanski, E.J. Park, R.S. McDowell, J.E. Theaker, N.J. Skelton, J. Am. Chem. Soc. 123 (2001) 625.
[17] P.D. Kwong, R. Wyatt, J. Robinson, R.W. Sweet, J. Sodroski, W.A. Hendrickson, Nature 393 (1998) 648.
[18] C. Das, S. Raghothama, P. Balaram, J. Am. Chem. Soc. 120 (1998) 5812.
[19] B. Odaet, F. Jean, C. Boutillon, E. Buissine, O. Melynk, A. Tartar, G. Lippens, Protein Sci. 8 (1999) 2773.
[20] E. Alba, A. Jimenez, M. Rico, J. Am. Chem. Soc. 119 (1997) 175.
[21] V. Munoz, L. Serrano, Proteins 20 (1994) 301.

[22] T.P. Quinn, N.B. Tweedy, R.W. Williams, J.S. Richardson, D.C. Richardson, Proc. Natl. Acad. Sci. USA 91 (1994) 8747.
[23] B.L. Sibanda, J.M. Thornton, Nature 316 (1985) 170.
[24] B.L. Sibanda, J.M. Thornton, J. Mol. Biol. 229 (1993) 428.
[25] M.S. Searle, J. Chem. Soc. Perkin Trans. 2 (2001) 1011.
[26] J.A. Robinson, Synlett 4 (1999) 429.
[27] J.S. Nowick, E.M. Smith, M. Pairish, Chem. Soc. Rev. (1996) 401.
[28] M. Erdelyi, V. Langer, A. Karlen, A. Gogoll, New J. Chem. 26 (2002) 834.
[29] S.J. Russel, A.G. Cochran, J. Am. Chem. Soc. 123 (2000) 625.
[30] D.L. Minor Jr., S.P. Kim, Nature 367 (1994) 660.
[31] K. Smith, L. Regan, Acc. Chem. Res. 30 (1997) 153.
[32] T.S. Haque, J.C. Little, S.H. Gellman, J. Am. Chem. Soc. 118 (1996) 6975.
[33] J.S. Nowick, Acc. Chem. Res. 32 (1992) 287.
[34] Y.J. Chung, B.R. Huck, L.A. Christianson, H.E. Stanger, S. Krauthauser, D.R. Powell, S.H. Gellman, J. Am. Chem. Soc. 122 (2000) 3995.
[35] A.G. Street, S.L. Mayo, Proc. Natl. Acad. Sci. USA 96 (1999) 9074.
[36] K.L. Constantine, L. Mueller, N.H. Andersen, H. Tong, C.F. Wandler, M.S. Friedrichs, R.E. Bruccoleri, J. Am. Chem. Soc. 117 (1995) 10841.
[37] M.S. Friadrichs, T.R. Stouch, R.E. Bruccoleri, L. Mueller, K.L. Constantine, J. Am. Chem. Soc. 117 (1995) 10855.
[38] E. Gallo, S.H. Gellman, J. Am. Chem. Soc. 116 (1994) 11560.
[39] M.S. Searle, S.R. Griffith-Jones, H. Skinner Smith, J. Am. Chem. Soc. 121 (1999) 11615.
[40] I.L. Karle, S.K. Awasthi, P. Balaram, Proc. Natl. Acad. Sci. USA 93 (1996) 8189.
[41] H.E. Stranger, S.H. Gellman, J. Am. Chem. Soc. 120 (1998) 4236.
[42] J.F. Espinosa, S.H. Gellman, Angew. Chem. Int. Ed. 39 (2000) 2420.
[43] I.L. Karle, H.N. Gopi, P. Balaram, Proc. Natl. Acad. Sci. USA 98 (2001) 3716.
[44] E. de Alba, M.A. Jimenez, M. Rico, J. Am. Chem. Soc. 119 (1997) 175.
[45] G.J. Sharman, M.S. Searle, J. Am. Chem. Soc. 120 (1998) 4869.
[46] T. Kortemme, M. Ramirez Alvarado, L. Serrano, Science 281 (1998) 253.
[47] S. Aravinda, N. Shamala, R. Rajkishore, H.N. Gopi, P. Balaram, Angew. Chem. Int. Ed. 39 (2002) 2330.
[48] M.H.V. Ramana Rao, S. Kiran Kumar, A.C. Kunwar, Tetrahedron Lett. 44 (2003) 7369.
[49] G. Srinivasulu, M.H.V. Ramana Rao, S. Kiran Kumar, A.C. Kunwar, Arkivoc (2004) 69.
[50] J. Venkatraman, S.C. Shankaramma, P. Balaram, Chem. Rev. 101 (2001) 3131.
[51] R. Kaul, P. Balaram, Bioorg. Med. Chem. 7 (1999) 105.
[52] A. Dutt, M.G.B. Drew, A. Pramanik, Tetrahedron 64 (2008) 549.
[53] A. Dutt, A. Dutta, R. Mondal, E.C. Spencer, A.J.K. Howard, A. Pramanik, Tetrahedron 63 (2007) 10282.
[54] A. Dutt, M.G.B. Drew, A. Pramanik, Tetrahedron 61 (2005) 11163.
[55] A. Dutt, R. Fröhlich, A. Pramanik, Org. Biomol. Chem. 3 (2005) 661.
[56] I.L. Karle, P. Balaram, Biochemistry 29 (1990) 6747.
[57] C. Toniolo, M. Crisma, F. Formaggio, C. Peggion, Biopolymers (Pept. Sci.) 60 (2001) 396.
[58] M. Bodanszky, A. Bodanszky, The Practice of Peptide Synthesis, Springer, New York, 1984, pp. 1–282.
[59] Crysalis, v1Oxford Diffraction Ltd., Oxford, UK, 2005.
[60] W. Kabsch, J. Appl. Cryst. 21 (1988) 916.
[61] G.M. Sheldrick, Acta Crystallogr. Sect. A: Fundam. Crystallogr. A46 (1990) 467.
[62] Oxford Diffraction Ltd., Abspack Program, 2005.
[63] A.L. Spek, PLATON Program, Utrecht University, The Netherlands, 2004.
[64] D.S. Wishart, B.L. Sykes, F.M. Richards, J. Mol. Biol. 222 (1992) 311.
[65] K. Wüthrich, NMR of Proteins and Nucleic Acids, Wiley, New York, 1986.