Configurationally guided peptide conformational motifs: crystal structure of a $L^{\alpha}D^{\beta}L^{\beta}D^{\alpha}D^{\beta}L^{\alpha}$ type hexapeptide fold

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A new concept in macromolecular design is described, using sequence configuration as the tool to control the topology of a compact polymer chain fold.

Residue configuration as a sequence variable provides a promising tool for the design of novel peptide conformational motifs. Illustrating the concept, we describe here a configurationally guided $L^{\alpha}D^{\beta}L^{\beta}D^{\alpha}D^{\beta}L^{\alpha}$ type peptide conformational motif. Residue preferences in the Ramachandran diagram[1] remain fundamental to the stereochemical outcome of peptide chain folding. Genetically encodable poly-L-configurational peptides are required in forming the familiar α-helix and β-sheet motifs,[2] because every residue needs to occur in the same left half of the Ramachandran diagram. The alternating L,D-configurational gramicidin A forms β-helical channel or β-double helical pore motifs,[3,4] while the alternating L,D-configurational cyclic peptides of Ghadiri and co-workers are self-organizing nanotubular structures,[5] because consecutive residues occur in the opposite halves of the Ramachandran diagram [see Fig. 1(*a*)]. The type II and type II' β-hairpins, being $L^{\beta}L^{\beta}D^{\alpha}L^{\beta}$ and $L^{\beta}D^{\beta}L^{\alpha}L^{\beta}$ type motifs based on the positional preferences for L- or D-configurational residues and their conformational states,[6] are most reliably approached by using sequence configuration as the design strategy.[7–9] Control of peptide chain folding *via* sequence configuration is thus a promising tool to achieve the design of unnatural peptide conformational motifs.

The L- and D-configurational residues in the hexapeptide Boc-L-Leu[1]-D-Val[2]-L-Pro[3]-D-Asp[4](OMe)-D-Val[5]-L-Leu[6]-NHMe† are noted to occur in the preferred halves of the Ramachandran diagram [Fig. 1(*b*)]. The configurational sequence LDLDDL combines with the conformational sequence αββαβα to produce the $L^{\alpha}D^{\beta}L^{\beta}D^{\alpha}D^{\beta}L^{\alpha}$ type motif, which has a compact backbone fold and a specific pattern of interpeptide hydrogen bonds‡ (Fig. 2). In its architectural plan, the motif may either be recognized as a consecutive type II-II' β-turn anchoring an unusual $L^{\alpha}D^{\beta}$ type dipeptide fold, or as a type II' β-hairpin with D configurational arms bracketed by L configurational residues in α type conformational states. An almost ideal type II' β-turn has Val[5]-Leu[6] as its corner residues (ϕ = 72, ψ = −117° for D-Val[5] against the ideal values 60 and −120°, and −81 and −14° for L-Leu[6] against the ideal values −90 and 0°) and the $i + 3 \rightarrow i$ type hydrogen bond between the methylamide NH and the Asp[4] CO. An almost ideal type II β-turn has Pro[3]-Asp[4] as its corner residues (ϕ = −64, ψ = 129° for L-Pro[3] against the required −60 and 120 , and 72 and 19° for D-Asp[4] against the requirement 90 and 0°) and the $i + 3 \rightarrow i$ type hydrogen bond between the Val[2] CO and the Val[5] NH. The D-configurational valines in the first and fourth position of this β-turn element are in the β region of the right half of the Ramachandran diagram (ϕ = 126, ψ = −92° for Val[2] and 72 and −117° for Val[5]) and with an additional hydrogen bond between the Val[2] NH and the Val[5] CO. The segment Val-Pro-Asp(OMe)-Val is thus a $D^{\beta}L^{\beta}D^{\alpha}D^{\beta}$ type β-hairpin, being an enantiomer of the naturally occurring $L^{\beta}D^{\beta}L^{\alpha}L^{\beta}$ type hairpin with a type II' β-turn and L-configurational arms and a diastereoisomer of another naturally occurring but stereochemically less preferred $L^{\beta}L^{\beta}D^{\alpha}L^{\beta}$ type hairpin with a type II β-turn and L-configurational arms.[10] Leu[1] at the N-terminal end has α-helical type torsional angles (ϕ = −66, ψ = −45° against the ideal values −57 and −48°) and has its NH hydrogen bonded to the Val[5] CO, which also participates in a bifurcated hydrogen bond with the Val[2] NH.

The Val[2] and Val[5] side chains are in a stack-like arrangement, while the Leu[6] side chain and Boc group are in a parallel arrangement, together describing a molecular cleft which accommodates the side chain of an Asp[4](OMe) from another molecule, providing for intermolecular packing. The masking of the Asp carboxylate and consequent suppression of its

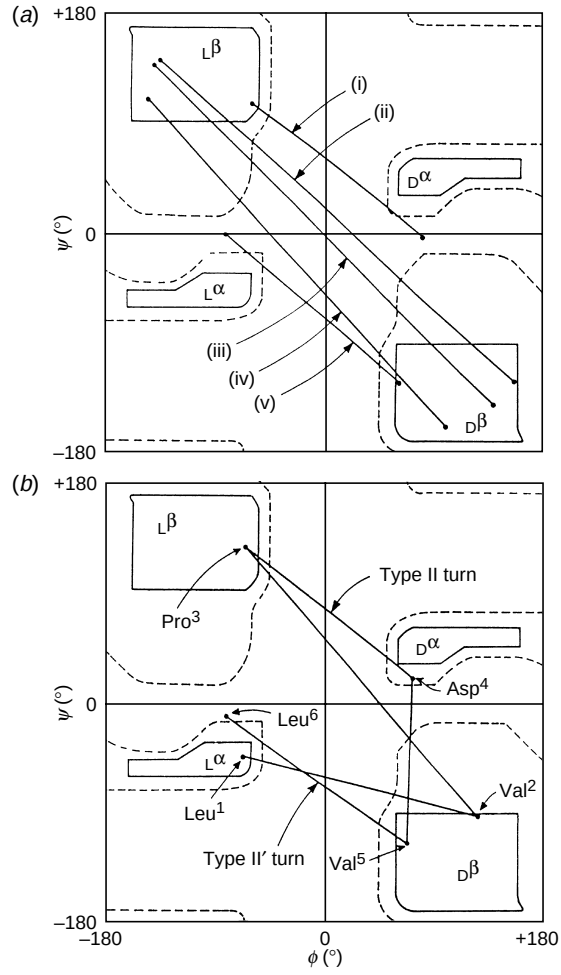

Fig. 1 The regions in the Ramachandran diagram favoured for L- and D-configurational residues. (*a*) The requirement of ϕ and ψ angles for (i) type II β-turn, (ii) gramicidin β-double helical pore, (iii) Ghadiri's octapeptide nanotubular structure, (iv) gramicidin β-helical channel and (v) type II' β-turn. (*b*) The ϕ and ψ angles in the hexapeptide Boc-L-Leu-D-Val-L-Pro-D-Asp(OMe)-D-Val-L-Leu-NHMe.

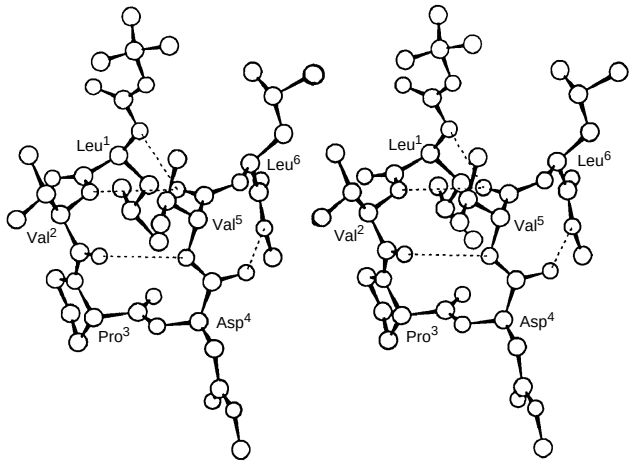

Fig. 2 Stereodiagram of the molecular structure of the hexapeptide Boc-L-Leu-D-Val-L-Pro-D-Asp(OMe)-D-Val-L-Leu-NHMe

tendency to hydrogen bond with a main chain NH is apparently a factor in the extended conformation of the Asp side chain and in its participation in this specific intermolecular packing arrangement. Intermolecular packing also features hydrogen bonds involving the Leu[6] NH and the CO group of Boc moiety from one molecule and, respectively, the Leu[1] CO and Asp[4] NH groups from two different molecules.

A most remarkable feature of the $L^{\alpha}D^{\beta}L^{\beta}D^{\alpha}D^{\beta}L^{\alpha}$ type motif is that its α-helix-favouring leucines occur in the α conformational region and its β-sheet-favouring valines occur in the β conformational region, while its turn-favouring Pro and Asp moieties occur in a β-turn element.[11] Thus the effects of structural origin, manifested in protein folding, are possibly operative over and above the effects of configurational origin, and will require consideration as part of the overall design strategy for the approach to other possible configurationally-guided peptide conformational motifs.

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Footnotes

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† The peptide synthesis was by solution phase methodology, the purification was by RP-HPLC on a C-18 column eluting with water–MeOH, and characterization was by NMR and mass spectrometry.

‡ Colourless rectangular crystals (0.75 × 0.15 × 0.2 mm) obtained from aq. MeOH were used for intensity data collection on an Enarf-Nonius CAD4 diffractometer with Cu-Kα (λ = 1.5418 Å) radiation. The crystals were monoclinic, space group $P2_1$, with cell parameters a = 10.059(1), b = 18.864(2), c = 12.586(1) Å; β = 91.62(5)°, V = 2383.9 Å, Z = 2, and diffracted up to 63°. The structure was solved using SHELXS-86 (ref. 12) and refined using SHELXS-93 (ref. 13). Hydrogens were refined as riding over the heavier atoms. The final R-factor was 0.046 (wR = 0.1254) for 3415 observed reflections with $F_o \geqslant 4\sigma(F_o)$. CCDC 182/504.

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