Abstract

Insertion of ω amino acid residues into all α polypeptide backbones leads to an expanded repertoire of secondary structures. In the present thesis an attempt has been made to explore the conformational properties of the γ amino acid residue gabapentin (1-aminomethylcyclohexaneacetic acid, Gpn). Gpn is a β,β′ disubstituted γ amino acid residue. The presence of a tetrasubstituted carbon atom in the backbone dramatically reduces the sterically allowed conformational space for gabapentin residues.

The introductory **Chapter 1** briefly summarizes the work that has been reported thus far on peptides containing ω amino acid residues. Gpn residues can adopt C_9 hydrogen bonded structures, a feature first observed in the crystal structures of Boc-$(Gpn)_2$-NHMe and Boc-$(Gpn)_4$-NHMe (Vasudev *et. al*., 2005).

Chapter 2, Section I presents a systematic study on terminally blocked oligomers of Gpn, Boc-(Gpn) n-NHMe, where n = 1-6. NMR studies establish C_9 ribbon conformations in solution for the n = 2 -6 cases in $CDCl_3$ solutions. The effect of inserting a guest Gpn residue into an all α amino acid host sequence is examined in **Chapter 2**, Section II. Three peptides Boc-^{L}Ala-Aib-Gpn-Aib-^{L}Ala-OMe (**1**), Boc-Leu-Aib-Val-Gpn-Leu-Aib-Val-OMe (**2**) and Boc-Leu-Aib-Val-Aib-Gpn-Aib-Leu-Aib-Val-OMe (**3**), all of which contain a centrally positioned Gpn residue are examined. Crystals of peptide **1** were obtained, wherein it formed a C_{12} helix stabilized by two C_{12} hydrogen bonds and one C_{10} hydrogen bond (Vasudev *et. al*., 2007). . Both peptides **2** and **3,** yielded clear NOE evidence in favor of helical conformations in solution. In all three peptides C_{12} hydrogen bonds were formed by the αγ/γα segments involving the guest Gpn residue. Isolated γ amino acid residues inserted into host α amino acid sequences, can be readily accommodated into helical folds, with an expansion of the hydrogen boded ring involving the guest amino acid residue.

Chapter 3 describes studies designated to probe the conformational preference of the alternating $(\alpha\gamma)_n$ hybrid sequences, where Gpn was used as the γ residue. When the α residue was Aib (α- aminoisobutyric acid), an α,α'- disubstituted constrained amino acid residue, $(\alpha\gamma)_n$ peptides adopted C_{12} helical conformations. Studies described in Chapter 3, establish a considerable decrease in the population of C_{12} helical conformations in solution and increased conformational heterogeneity, when Aib residues are replaced by Ala.

In Chapter 4, αγ hybrid peptides with tripeptide repeat segments $(\alpha\gamma\alpha)_n$ and $(\gamma\alpha\alpha)_n$ have been studied. A helical conformation was characterized in crystals for the peptide Boc-Leu-Gpn-Aib-Leu-Gpn-Aib-OMe (**1**) (Chatterjee *et. al*., 2008b). For the peptide **1**, there was evidence of conformational exchange in solution, with the observation of medium range NOEs which were incompatible with the helical conformation characterized in crystals for

1. Observation of a single set of NH resonances in the NMR spectrum, is attributed to rapid rates of interconversion between the different conformational species. For peptides Boc-Gpn-Leu-Aib-Gpn-Leu-Aib-OMe (**5**) and Boc-Gpn-Val-Aib-Gpn-Val-Aib-OMe (**6**), with $(\gamma\alpha\alpha)_n$ tripeptide repeats, conformational studies were performed in $CDCl_3$ solution using NMR methods. Two sets of sharp NH resonances of equal intensity were observed in the spectrum at ambient temperature. This was indicative of a slow rate of exchange between conformers which were of similar thermodynamic stability.

Chapter 5, presents a novel 12/10 helical structure established in the crystalline state for the peptide Boc-Leu-Gpn-Leu-Aib-OMe...The Leu (1)-Gpn (2) segment forms a $4\rightarrow1$ C_{12} hydrogen bond with the normal directionality (CO(i)…NH (i+3)). A second C_{10} hydrogen bonding is formed between the Gpn (2) NH and Leu (3) CO ($1\rightarrow2$), in which the hydrogen bond directionality is opposite (NH(i)…CO (i+1)). Both Leu residues adopt P_{II} conformations (ϕ = -60° and ψ = 120°), while the Gpn residue adopts a *gauche-gauche* conformation with ϕ = 87°, θ_1 = 38°, θ_2 = 45° and ψ = -129° (Vasudev *et. al*., 2008)

Chapter 6 addresses the question of the effect of inserting Gpn into sequences which have been shown to form both helices and hairpins. X-ray diffraction studies revealed a β-hairpin conformation of peptide Boc-Leu-Phe-Val-Aib-Gpn-Leu-Phe-Val-OMe in crystals, nucleated by a C_{12} hydrogen bond across the Aib-Gpn segment. This structure provides the only example of a Gpn residue in a *gauche-trans* (*gt)* conformation with a value of $\theta_2 \sim 180°$. In $CDCl_3$ solutions, the peptide predominantly adopted a helical conformation, while in CD_3OH, the peptide favoured a hairpin conformation as characterized in crystals. This solvent dependent conformational transition together with the presence of minor conformations in solution, indicates that the Aib-Gpn segment is not a robust β-hairpin nucleating structural feature. The studies described in this thesis, establish that the Gpn residue is a versatile, stereo chemically constrained γ amino acid residue, which can be usefully employed in the design of hybrid peptides with well defined backbone conformations.