Investigation of Schellman Loops

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This abstract presents the results of an analysis of Schellman loops in proteins and polypeptides. A further aim of this work was to investigate a possible structural resemblance between these loop features and motifs formed by the interaction between asp, asn, ser and thr sidechain oxygens with nearby mainchain amides.

Analysis of the work of Deane [1], Duddy [2] and Wan [3,4] raises the possibility that many of the contacts between the asx- and ST- sidechain oxygen atoms and the nearby amide hydrogen could be occurring within motifs called ß-bulge loops having the first residue as either D, N, S or T. Based on this, I investigated their geometry.

Schellman loops, also called Paperclips [5-7], are commonly occurring six to seven residue motifs incorporating a nest [8]. There are two types of Schellman loop according to the number of residues: a classic type which consists of six residues and has two characteristic hydrogen bonds. The other type is called a wide Schellman loop. It consists of seven residues and has also two hydrogen bonds.

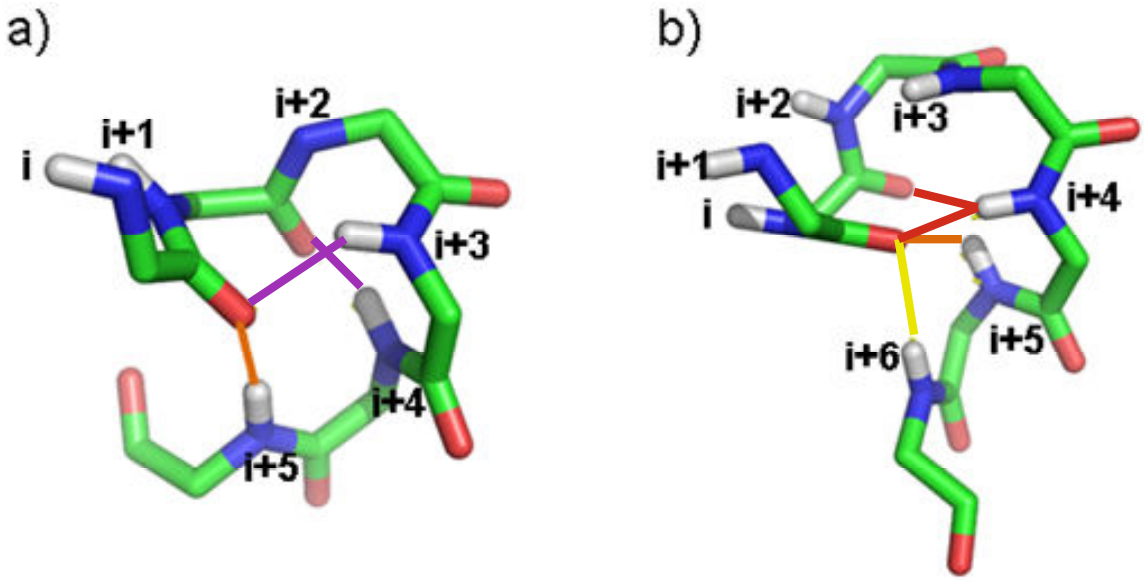

Figure 1. Classic and wide RL Schellman loops. Stick displays using Pymol with the amino acid residues labelled at their NH groups, for a) the classic RL Schellman loop and b) the wide RL Schellman loop.

A database of 500 protein high resolution pdb structures [9] was searched for the different types of Schellman loops. The search was performed according to specific criteria as the following steps:

1. Search for six (or seven) successive residues.

2. One hydrogen bond was defined between the oxygen of the carbonyl group in residue i and the hydrogen of the amide group of residue i+5 (or i+6).

3. Another hydrogen bond between the oxygen of the carbonyl group in residue i+1 and the hydrogen of the amide group of residue i+4 (or i+5).

4. Residues i+3 (or i+4) have αR conformations.

5. Residues i+4 (or i+5) have αL conformations.

6. To define hydrogen bonds:

    a. The O….H distance should be ≤ 2.5Å.

b. The C-O….H angle is ≥ 90°.

c. The O….H-N angle is ≥ 90°.

In the 500 protein database, 862 unique classic Schellman loops were found. In classic Schellman loops, my studies have revealed a phenomenon linking the two hydrogen bonds (i-1, i+3) and (i, i+3). They have the interesting property that, when (i-1, i+3) is strong, (i, i+3) is weaker. When (i, i+3) is stronger, (i-1, i+3) is weaker. The two carbonyl oxygens appear to compete for the one NH group. This competition can be seen in Figure 2a for the O…H distances and in Figure 2c for the O…H-N angles. Much less compensation is seen for the C-O…H angle in Figure 2b.

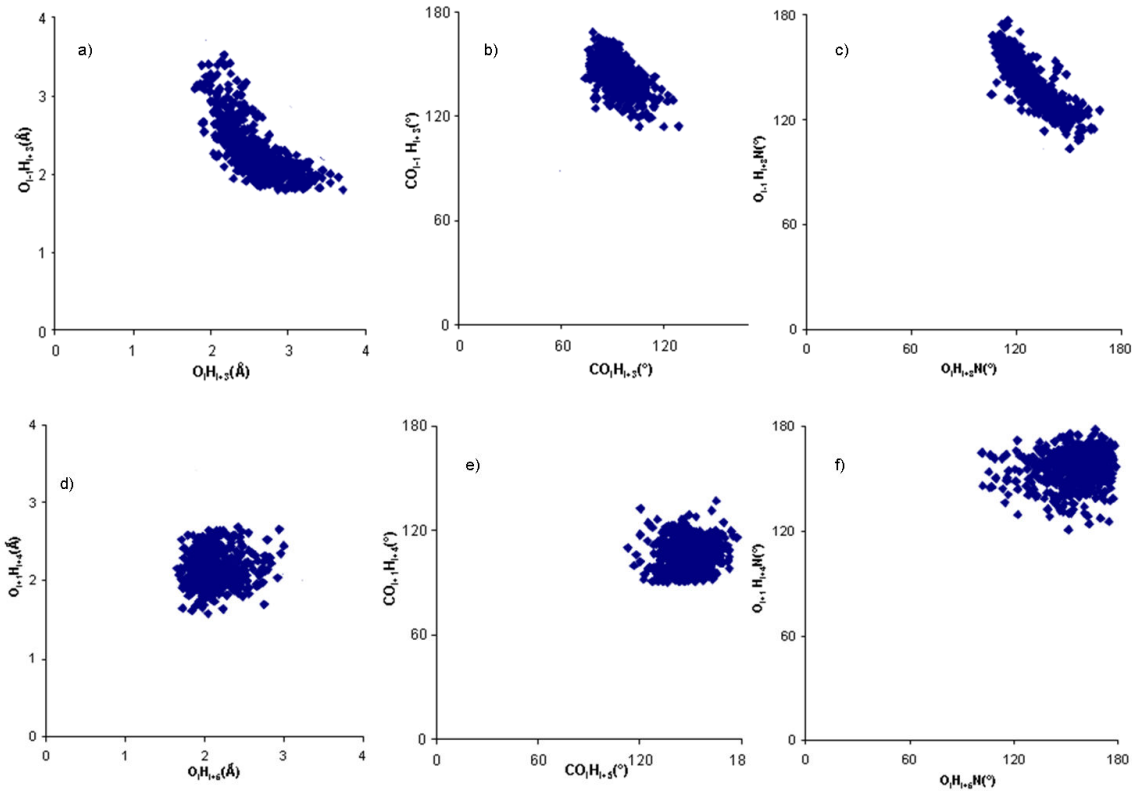

Figure 2. Evidence for Competing and Non-Competing Hydrogen Bonds in the Schellman Loop. The Competing ones in the top figures are O_{i-1}- H_{i+3} (y-axis) and O_i - H_{i+3} (x-axis). The Non-Competing ones in the bottom figures are O_i - H_{i+4} (y-axis) and O_{i+1}- H_{i+5} (x-axis). a) and

d) O-H distance comparisons. b) and e) COH angle comparisons. c) and f) OHN angle comparisons.

Figure 2, reveals that the strength of the (i, i+3) hydrogen bond in Schellman loops is variable and that it hardly ever becomes as strong as the (i, i+5) bond. Thus, although the nest is often portrayed as chelating one anion by the first and the third mainchain NH groups, in Schellman loops the two NH groups often bind different oxygen atoms. Nests, type I β-turns and π-turns are integral to all classic Schellman loops, while some other motifs are liable to be incorporated in Schellman loops including, asx-turns, ST-turns, asx-motifs, ST-motifs and rarely niches.

Typically, type I and type II β-bulge loops possess of an asp, asn, ser or thr at residue i. In the Richardson Database of 500 proteins there are: 394 β-bulge loops, 244 of type I and 150 of type II. Seventy three percent of the type I β-bulge loops have an asp, asn, ser or thr as the first residue of the motif; of these 65% are either asp or asn. Beta-bulge loops with the first residue as either asp or asn commonly exhibit a structural resemblance, via sidechain-mainchain mimicry by asp, asn, ser or thr, to RL classic Schellman loops, as seen in Figure 3.

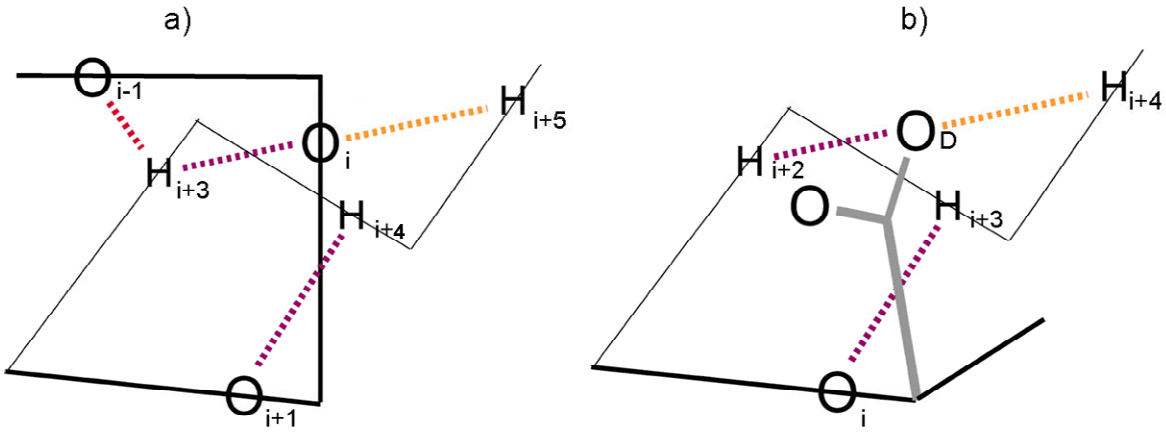

Figure 3. **. Diagrammatic representation of the two motifs**. a) Shows a classic Schellman loop, while b) shows a type I ß-bulge loop with an aspartate side chain of residue i mimicking the main chain of residue i of the Schellman loop. CO_i -> NH_{i+3}, CO_i -> NH_{i+5}, CO_{i-1} -> NH_{i+3} and CO_{i+1} -> NH_{i+4} hydrogen bonds are shown as dashed lines and coloured magenta, orange, red and magenta respectively.

Superimposition on computer was performed, using Pymol, with nineteen atoms from the critical hydrogen bonded loops of both types of motif. The results show

that the two types of motifs are superimposable giving rise to an RMS fit around 0.4 Å as shown in Figure 4. An explanation for the mimicry by the ß-bulge loop is that the anion-binding propensity of the RL nest is satisfied by binding to two carbonyl oxygens in the same way in both motifs.

The majority of type I ß-bulge loops where the first residue is asp, asn, ser or thr mimic classic Schellman loops are called asp-, asn-, ser- and thr-ß-bulge loops. All thr-ß-bulge loops are mimics while 88% of ser- , 86% of asp- and 78% of asn-ß-bulge loops mimic classic Schellman loops.

The results indicate that the parameters of the strong bond are very similar (The variability of the torsion angle being merely a reflection of the CO…HN linearity). On the other hand (i, i+3) bond, known to be weaker in the Schellman loop, is less weak in all its key parameters (a distance and two angles) in the mimic, approaching, but not attaining, the strength of the other bond.

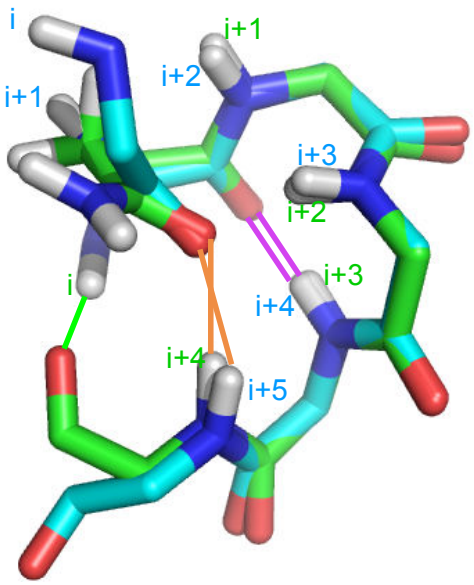

Figure 4. Superimposition of Schellman loop and its mimic. The top (blue) model is a classic Schellman loop (1csl) residues 111-116. The bottom (green)model is a ß-bulge loop (1cem) residues 125-129, The first residue being asn.

Conclusions

Schellman loops are categorized into four different types depending on the amino acid number and conformation of the nest residues incorporated. The four possible types are as follows: RL Schellman loop classic type, LR Schellman loop classic type, RL Schellman loop wide type and LR Schellman loop wide type. The classic type consists of six residues and the nest residues are in the fourth or fifth positions. The wide types consist of seven residues and the nest residues are in the fifth and sixth positions.

I have found that there is competition between the two hydrogen bonds (i-1, i+3) and (i, i+3). They have an interesting property that when when (i-1, i+3) is strong, the other (i, i+3) is weaker. In other words, the NH of i+3 can bind to the CO of i-1 and i, but when one bond is strong the other is weak. The (i, i+3) hydrogen bond is almost always weaker than (i, i+5), but it is also variable in its strength and is weakest when a stronger (i, i+5) bond is present. There appears to be a continuous distribution of energies of the two hydrogen bonds such that the situation with half of (i, i+3) and half of (i, i+5) occurs just as often as that

with mainly one bond or the other. However, the electron density in crystal structures need not necessarily reflect reality in that the situation may be an equilibrium between the two hydrogen bonds, with either one or the other, but not both. Averaging may then generates electron density in between. There are many examples from the chemical studies of small molecules where it is difficult to distinguish between fluctuating hydrogen bonds [10].

Comparison of four parameters in both Schellman loops and their mimics taking into account two different hydrogen bonds (i, i+3) and (i, i+5) indicates variability between the two motifs. The (i, i+3) parameters: O…H distance, O…HN angle, CO…HN torsion angle and CO…H angle differ in both motifs being stronger and invariable in the mimics (ß-bulge loops) compared to being weaker and variable in the Schellman loops.

Examination of the hydrogen bond parameters in type I ß-bulge loops, Schellman loop mimics and Schellman loops themselves confirms the homology between them. The two prospective hydrogen bond distances O…H in three different hydrogen bonds (i, i+2 or i+3), (i, i+4 or i+5) and (i+1, i+3 or i+4) reveals this structural resemblance in both motifs.

An interesting phenomenon linking the two hydrogen bonds between residues i-1, i and i+3 is that, when that between i-1 and i+3 is strong, that between i and i+3 is weak. There is a continuous distribution of conformations between these two extremes rather than one of the bonds always being stronger than the other. This probably reflects continual motion within the motif between two shallow energy minima.

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