# Buried β–turns in Hydrophobic Cores: Structural and Functional Implications

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## Abstract

A turn is a protein secondary structure where the polypeptide chain reverses its direction. In particular, a β–turn consists of only four consecutive residues and a γ–turn, which is the second most commonly found turn after β–turns, involves three consecutive residues. Turns are usually located at the molecular surface. However, they can sporadically be found buried within the protein hydrophobic core and the precise role of such occurrences remains unknown. In this study, we identify such buried turns and we examine putative structural and functional implications of their presence. In order to search for these patterns, we developed a program which incorporates and systematically applies PROMOTIF's and Stride's algorithms and identifies buried turns in a large sample of structures obtained from the Protein Data Bank via the PISCES interface. Preliminary results obtained from this procedure are presented.

## Introduction

A β–turn is a secondary structure motif which involves four consecutive residues, $R_i$, $R_{i+1}$, $R_{i+2}$, $R_{i+3}$, where the distance between the atoms $Ca_i$ and $Ca_{i+3}$ is less than 7 Å and the two central residues are not in a helical conformation. β–turns are the most commonly found turns and are classified into nine different types (Type I, Type I', Type II, Type II', Type IV, Type VIa1, Type VIa2, Type VIb, Type VIII) using the ϕ and ψ angles of residues $R_i$ and $R_{i+2}$[1]. The second most commonly found turns are γ–turns. A γ–turn consists of three consecutive residues, $R_i$, $R_{i+1}$, $R_{i+2}$, with a hydrogen bond forming between the backbone $CO_i$ and $NH_{i+2}$. γ–turns are divided into two different types, classic and inverse. The main chain atoms of the two forms can be found in two possible enantiomers. Turns are more frequently found on the solvent–exposed surface of the proteins, however, there have been reports where turns have been located buried within the hydrophobic core of the protein[2]. The precise structural and functional significance of such occurrences is currently unknown. Here we identify such structures in a large sample of structures obtained from the PDB, and we attempt to identify common structural and functional features shared between them.

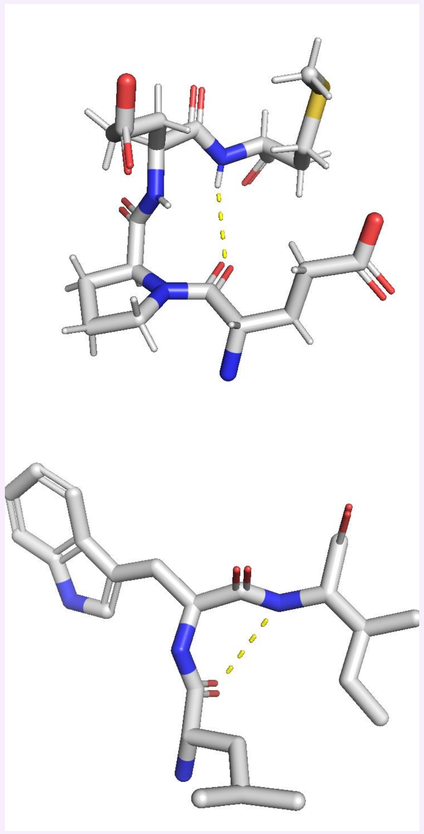


Figure 1. A β- and a γ-turn

## Methods

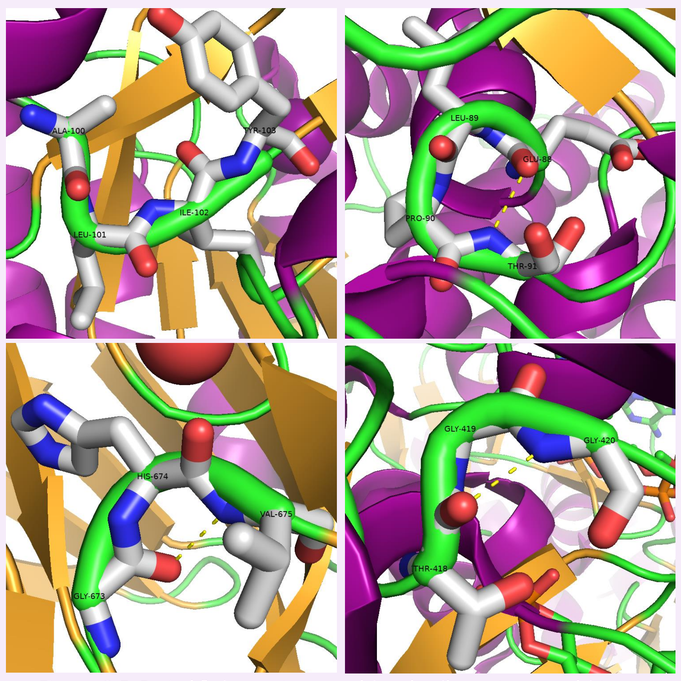
- 24951 protein entries were downloaded from the PDB with a list obtained from the culling server PISCES[3] with the criteria: maximum 2.2 Å resolution, maximum R-factor 1.0 and 70% identity cut–off
- PROMOTIF[4] was used for the identification of β- or γ-turns and STRIDE[5] was used for the calculation of solvent accessibility of the residues involved in the turns
- Gnuplot was used for the plotting of the density distributions and REVIGO[6] for the similarity-based scatterplot
- Pictures showing the hydrophobic buried turns were created using PyMOL[7]
- All computational scripts are written using the Perl programming language

## Results

- A total of 427 β– and γ–turns with a maximum solvent accessibility number of the residues involved equal to 0.0 were found. These 427 turns involve a total of 397 proteins out of the 24921 proteins in the initial sample, i.e., an average of 1.6% of protein structures included one or more hydrophobic turns.
- 88% of the proteins that include hydrophobic turns have catalytic properties. In particular, 45% of the proteins found belong to the hydrolase protein family, 15% in the oxidoreductase, 13% in the transferase and 7% in the lyase.
- 60% of the turns found are β-turns and 40% γ-turns.
- The most common turn type for buried β-turns is type IV (56%), followed by type I (15%) and type VIII (14%) and for buried γ-turns the inverse type (96%).
- The histograms plotted reveal a peak at 0.0, a drop at around 1.0, another peak at around 2.0, a plateau from 2.0 till 60.0 and a greater peak around 100.0.

| Classification | Hydrolase | Oxidoreductase | Transferase | Lyase | Isomerase | Unknown Function | Transport Protein | Ligase | Sugar Binding Protein | Structural Protein | Viral Protein | Immune System | Cell Adhesion | Metal Binding Protein | Membrane Protein | Transcription | Other Protein Families |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Number | 178 | 58 | 51 | 26 | 14 | 14 | 11 | 8 | 6 | 4 | 4 | 4 | 3 | 3 | 2 | 2 | 13 |

Table 1. Number of proteins in each protein family




Figures 2-5. Buried β- (upper two panels) and γ- (lower two panels) turns

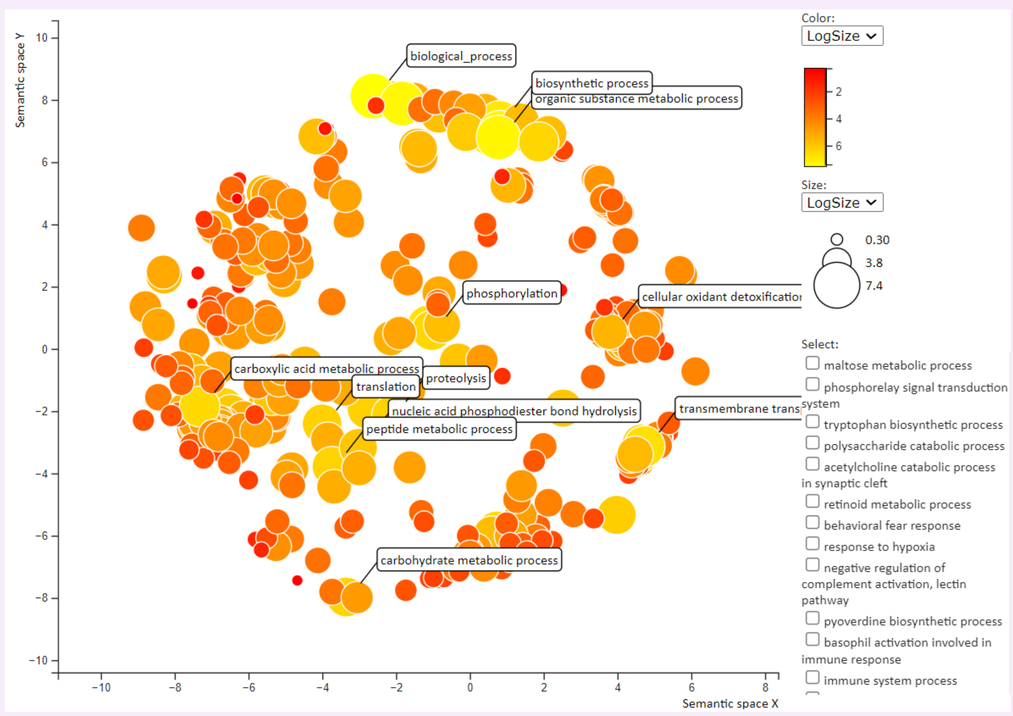



Figure 6. Similarity-based scatterplot

| Turn Type per Turn | | | |
|---|---|---|---|
| β-turns | | γ-turns | |
| I | 39 | CLASSIC | 7 |
| I' | 4 | INVERSE | 163 |
| II | 21 | | |
| II' | 8 | | |
| IV | 144 | | |
| VIa1 | 1 | | |
| VIa2 | 0 | | |
| VIb | 3 | | |
| VIII | 37 | | |

Table 2. Turn type per turn frequency

## Conclusion

To conclude, a significant number of turns buried inside the hydrophobic cores of a non-redundant set of proteins have been identified. Hydrophobic β-turns were found slightly more frequently than hydrophobic γ-turns, with the predominant type for β-turns being the miscellaneous IV type and the inverse type for γ-turns. Our results show that types IV and VIII are more prevalent in hydrophobic turns comparing to the general β-type frequency. Moreover, buried turns have been more frequently identified in proteins with enzymatic properties and, in particular, in proteins belonging in the hydrolase family. These results may be indicative of a preserved function associated with buried turns, however, further evaluation is necessary.

## Future Work

Our future intentions involve the construction of an RMSD or TM-score-based distance matrix with the aim of further reducing the structural redundancy that may be present in our data set. Furthermore, we will analyze our data in terms of the function of the proteins that we have identified with the aim of characterizing any putative functional significance of the buried turns. Additionally, we will analyze the structural context within which these turns are located with the aim of identifying any systematic trend in terms of their structural environment.

## References

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