

# A Study of Transmembrane Helix Distortions with Computational Tools

## Introduction

Structural irregularities are present in more than 70% of  $\alpha$ -helices in structural databases. One of the most frequent is the  $\pi$ -bulge, a transitional wide turn in the helix.

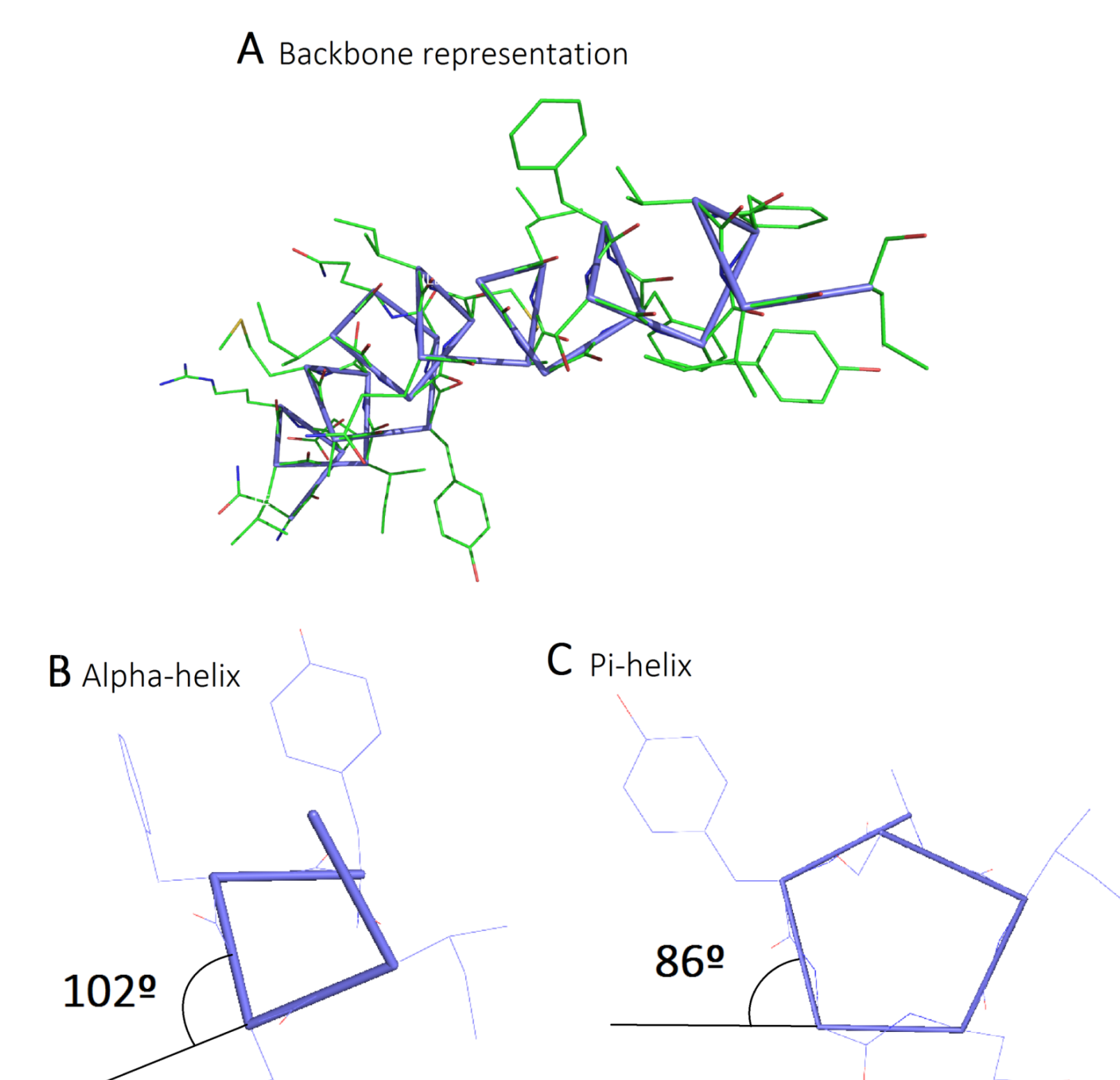
Many causes for this type of distortions have been described, such as ligand or residue interactions, presence of Pro and Gly turns and aminoacid insertions or deletions.

In this project a method to identify such distortions in TM helices is proposed. The results show three TM protein superfamilies with the described irregularities. Functional consequences of these  $\pi$ -bulges are discussed.

## Method

1. Extract the TM protein crystals from the database **TMalphaDB** ([lmc.uab.cat/TMalphaDB](http://lmc.uab.cat/TMalphaDB), 276 struc.)
2. Select the TM helix regions of the TM protein.
2. Calculate the backbone twist angles (Figure 1).
3. Plot the twist angles and identify the  $\pi$ -bulges.
4. Structural analysis with **PyMol** of the TM helices with  $\pi$ -bulges.
5. Structural alignment of the helices with their **Pfam** homologous ([pfam.xfam.org](http://pfam.xfam.org), 14831 fam.)
6. Identify the structurally conserved residues.
7. Sequence alignment of the TM helices compared, introducing the necessary gaps.

Figure 1. Helix conformations



## Results

### Cytochrome C oxidases

The cytochrome C oxidases (COX) have two  $\pi$ -bulges in its TM helix 2 (Figure 2B). One is the result of a Phe insertion, compared to the nitric oxide reductases (NOR). The ubiquinol oxidases (UOX) share this helix irregularity, but a later Gly deletion undoes the second COX's  $\pi$ -bulge (Figure 2C). The NOR's helix 2 is a canonical  $\alpha$ -helix (Figure 2D). The alignment of the Figure 2F shows the structurally conserved residues in black and the residue gaps in red.

### Ammonium Transporters

The Amt proteins are ammonium transporters, as the Rh proteins, and they are members of the same protein superfamily. However, a structural difference in its TM helix 4 is shown in the Figure 3A. There is a  $\pi$ -bulge in the Amt protein (Figure 3B), caused by a Phe insertion between the conserved residues Tyr and Leu. The alignment of the Figure 3E represents the structural findings made.

### Cation-Transporting ATPases

The cation pumps family also has members with the presented helix irregularity. The Na<sup>+</sup>/K<sup>+</sup>-transporting ATPases have a Gly insertion in the TM helix 7 (Figure 4B), compared to the SERCA pumps, which have a canonical  $\alpha$ -helix (Figure 4C). The alignment of the TM helices was more difficult in this particular case, because the SERCA TM helix 7 has a soft bend in the middle (Figure 4A).

Figure 2. Cytochrome C oxidases

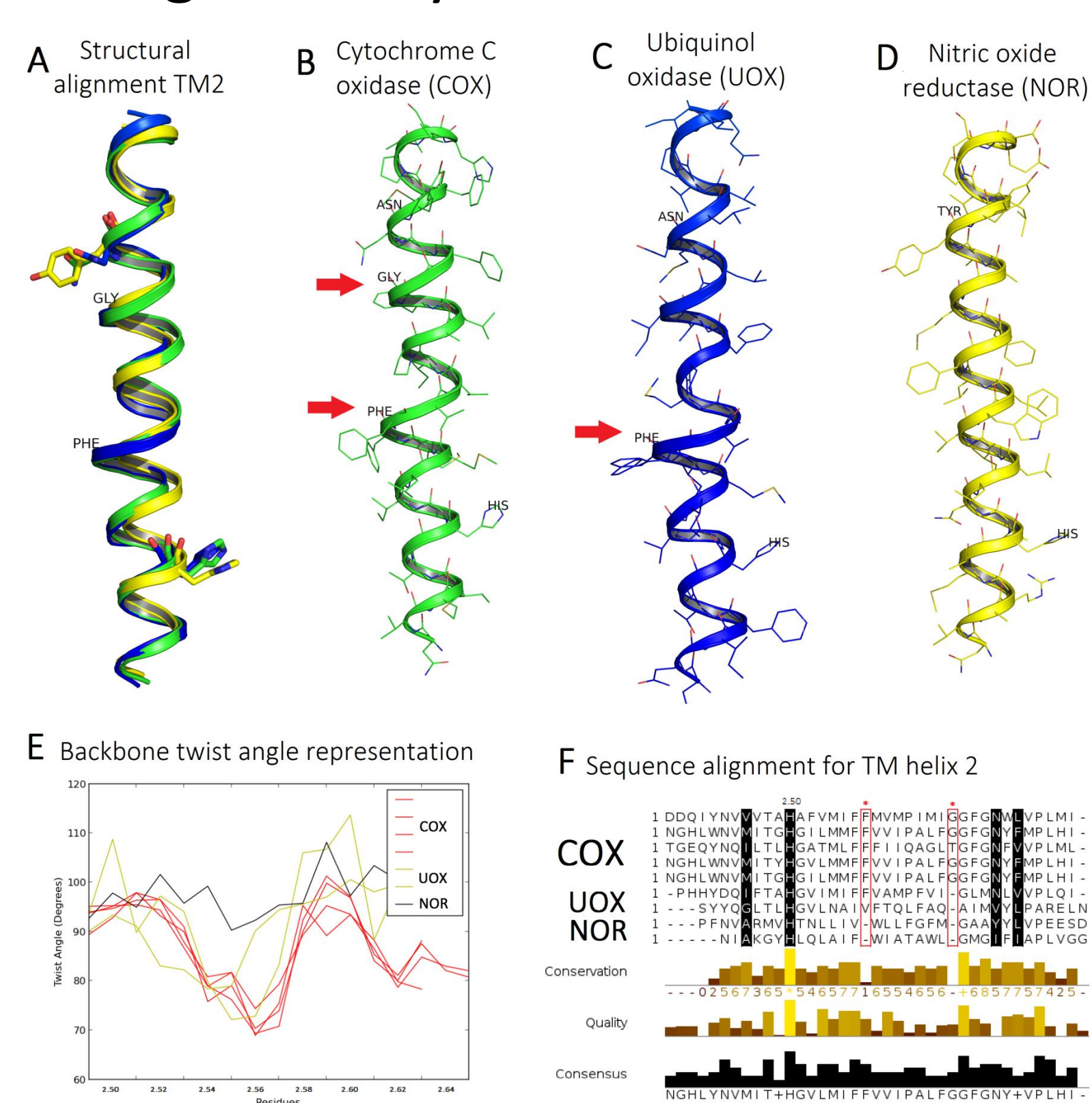


Figure 3. Ammonium Transporters

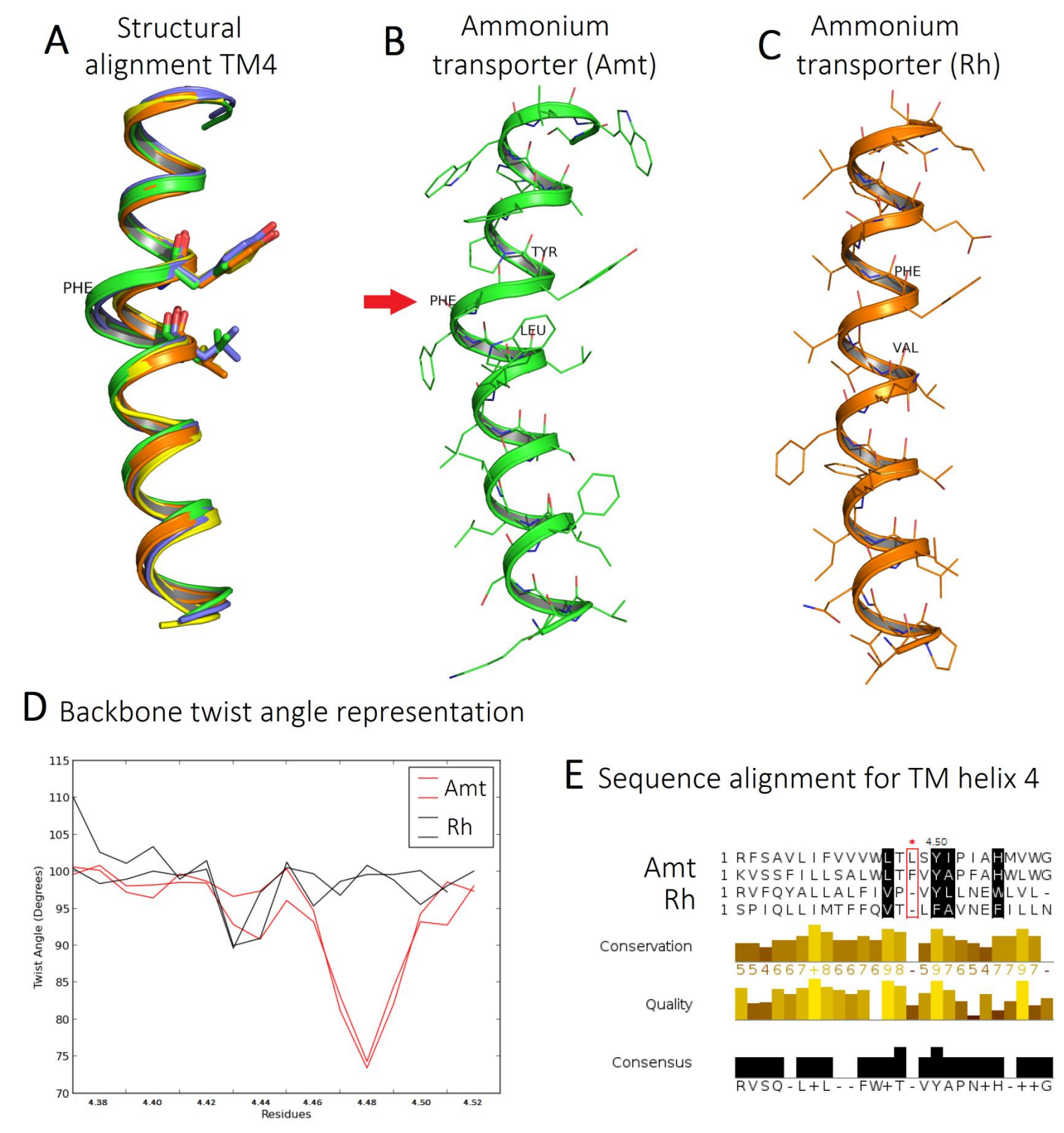
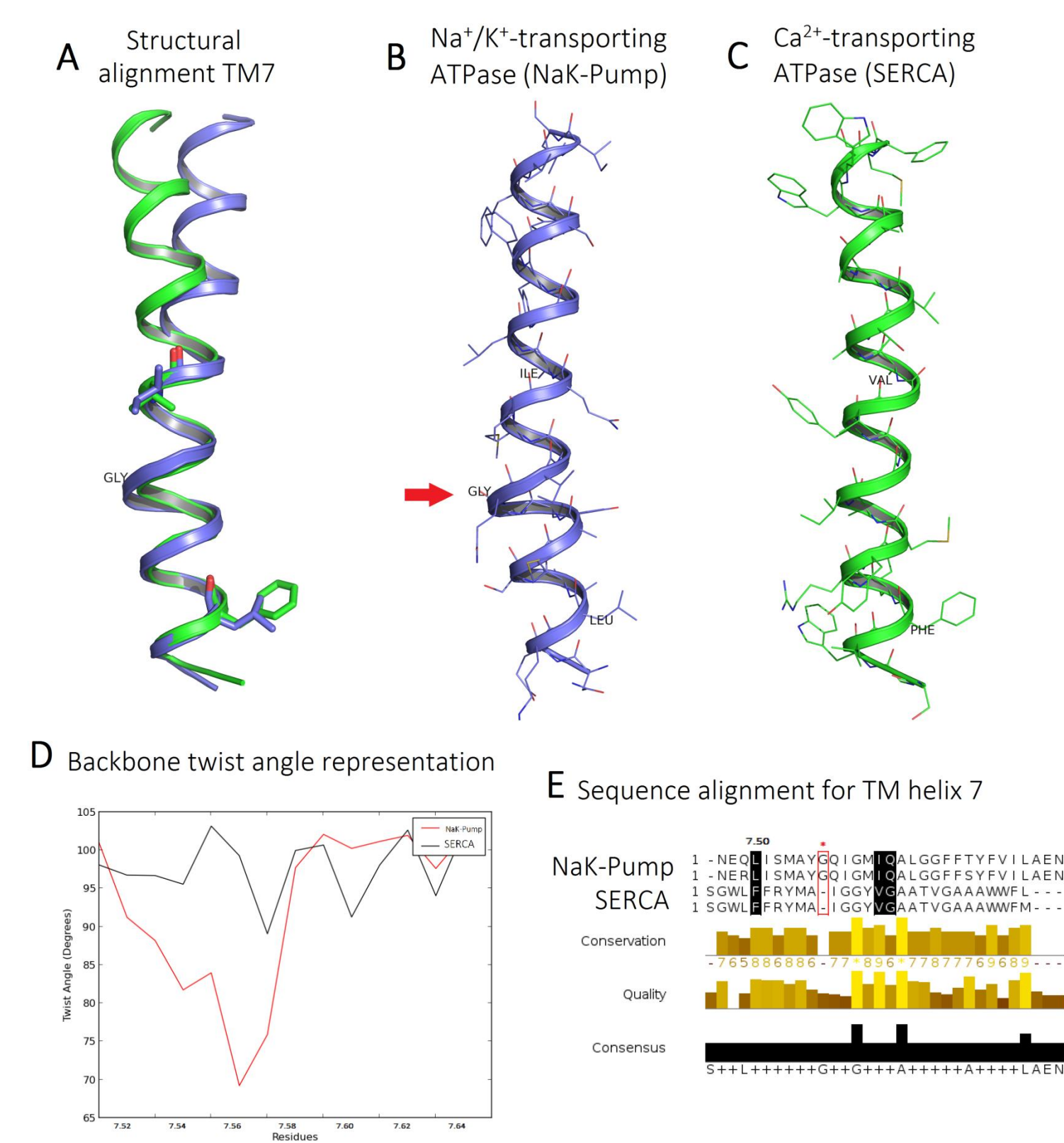


Figure 4. Cation-Transporting ATPases



## Research

The results of this project are only an early stage of all the research plan. Some families with helix irregularities have been identified, but many other structures need to be analyzed, following the method proposed, in order to establish an evolutionary path of such changes.

Another proposal is to analyze other helix irregularities than  $\pi$ -bulges, such as  $3_{10}$  helices or helix bends, in order to have an extended view of the functional consequences.

The final goal of this study is to find the reasons that lead to these changes in protein structure, in order to understand the functional consequences. A complementary proposal is to perform molecular dynamics studies of the helix irregularities found to study their interactions and deduce the possible functional consequences (Video).

## References

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