Course Project: Structural Analysis of the Hemoglobin Complex

Proteomes Interactomes and Biological Networks

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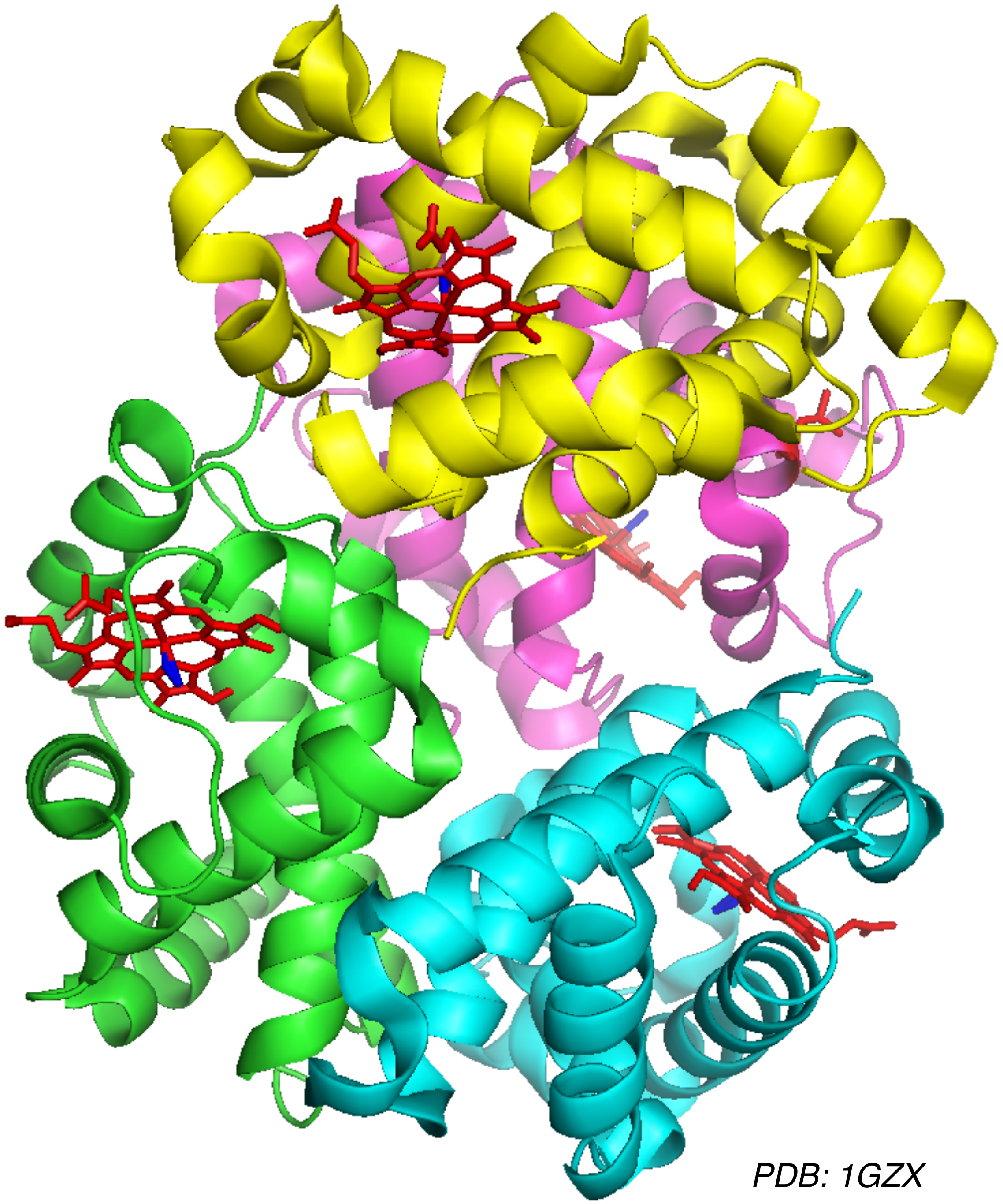
Hemoglobin

Hemoglobin is an oxygen-transport protein. It is a tetramer composed by two subunits designated α and β, with stoichiometry α2β2.

The four subunits of hemoglobin sit roughly at the corners of a tetrahedron, facing each other across a cavity at the center of the molecule. Each of the subunits contains a heme prosthetic group.

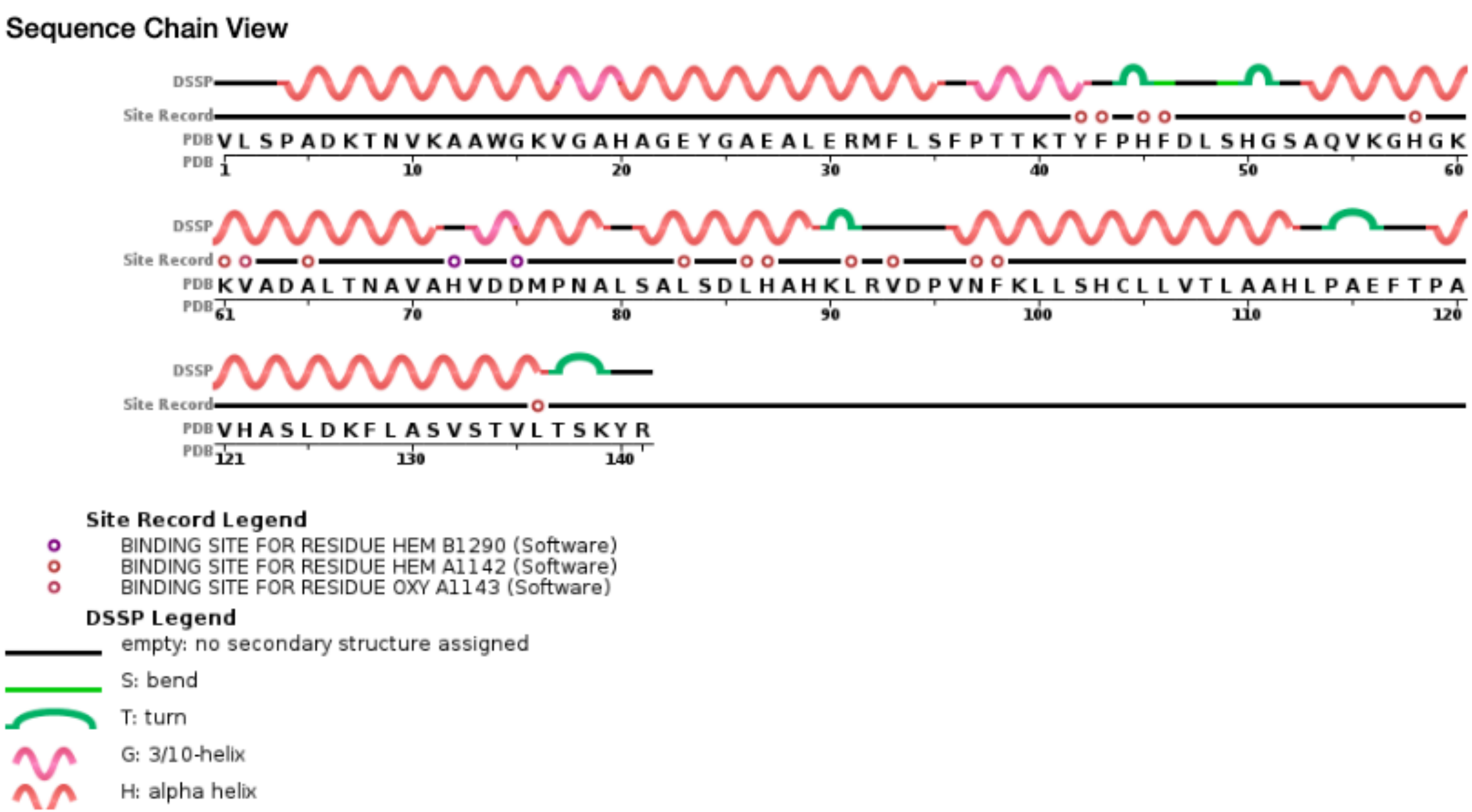
Each individual heme molecule contains one Fe2+ atom. The heme group binds oxygen while still attached to the hemoglobin monomer.



PDB: 1GZX

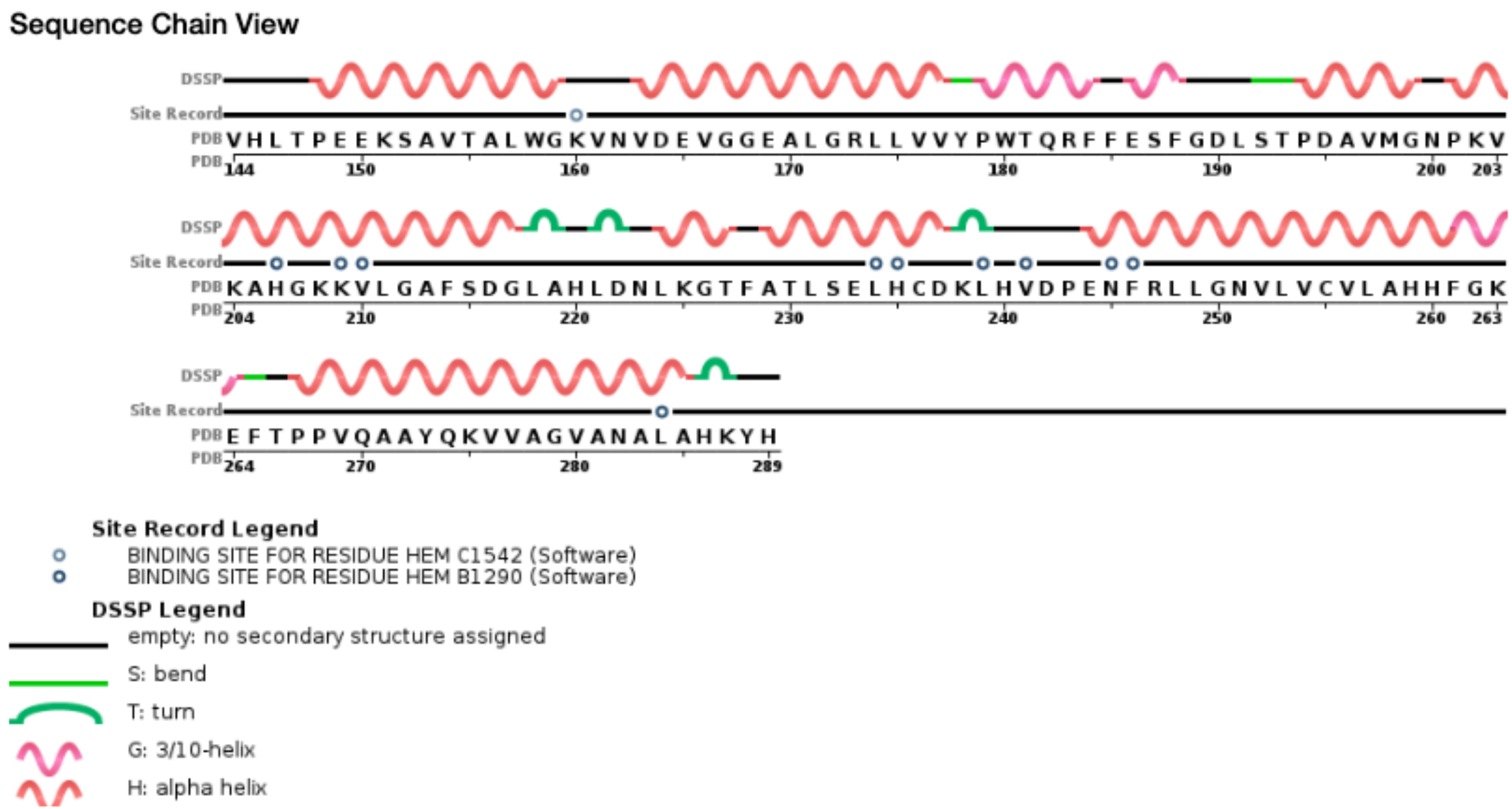
Subunit Alpha

All-alpha chain A and C of the PDB structure 1GZX composed by 141 residues



Subunit Beta

All-alpha chain B and D of the PDB structure 1GZX composed by 146 residues



Sequence Comparison

The alignment of the two sequences shows that they share ~44% of the residues

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The best scores are:                                      n-w bits E(1)
HBB_HUMAN 147 bp                                   ( 147)  373 65.3 1.4e-161

>>HBB_HUMAN 147 bp                                        (147 aa)
 n-w opt: 373  Z-score: 320.6  bits: 65.3 E(1): 1.4e-161
global/local score: 373; 43.6% identity (74.5% similar) in 149 aa overlap (1-142:1-147)

                10        20        30        40         50
HBA_HU MV-LSPADKTNVKAAWGKVGAHAGEYGAEALERMFLSFPTTKTYFPHF-DLS-----HGS
       :: :.: .:. : : ::::  .. : :.::: :... .: :. .:  : :::      :.
HBB_HU MVHLTPEEKSAVTALWGKV--NVDEVGGEALGRLLVVYPWTQRFFESFGDLSTPDAVMGN
               10          20        30        40        50

            60        70        80        90       100       110
HBA_HU AQVKGHGKKVADALTNAVAHVDDMPNALSALSDLHAHKLRVDPVNFKLLSHCLLVTLAAH
        .::.:::::  :.....::.:.. .....::.::  ::.::: ::.::.. :. .:: :
HBB_HU PKVKAHGKKVLGAFSDGLAHLDNLKGTFATLSELHCDKLHVDPENFRLLGNVLVCVLAHH
       60        70        80        90       100       110

           120       130       140
HBA_HU LPAEFTPAVHASLDKFLASVSTVLTSKYR
       .  :::: :.:. .: .:.:...:. ::.
HBB_HU FGKEFTPPVQAAYQKVVAGVANALAHKYH
      120       130       140
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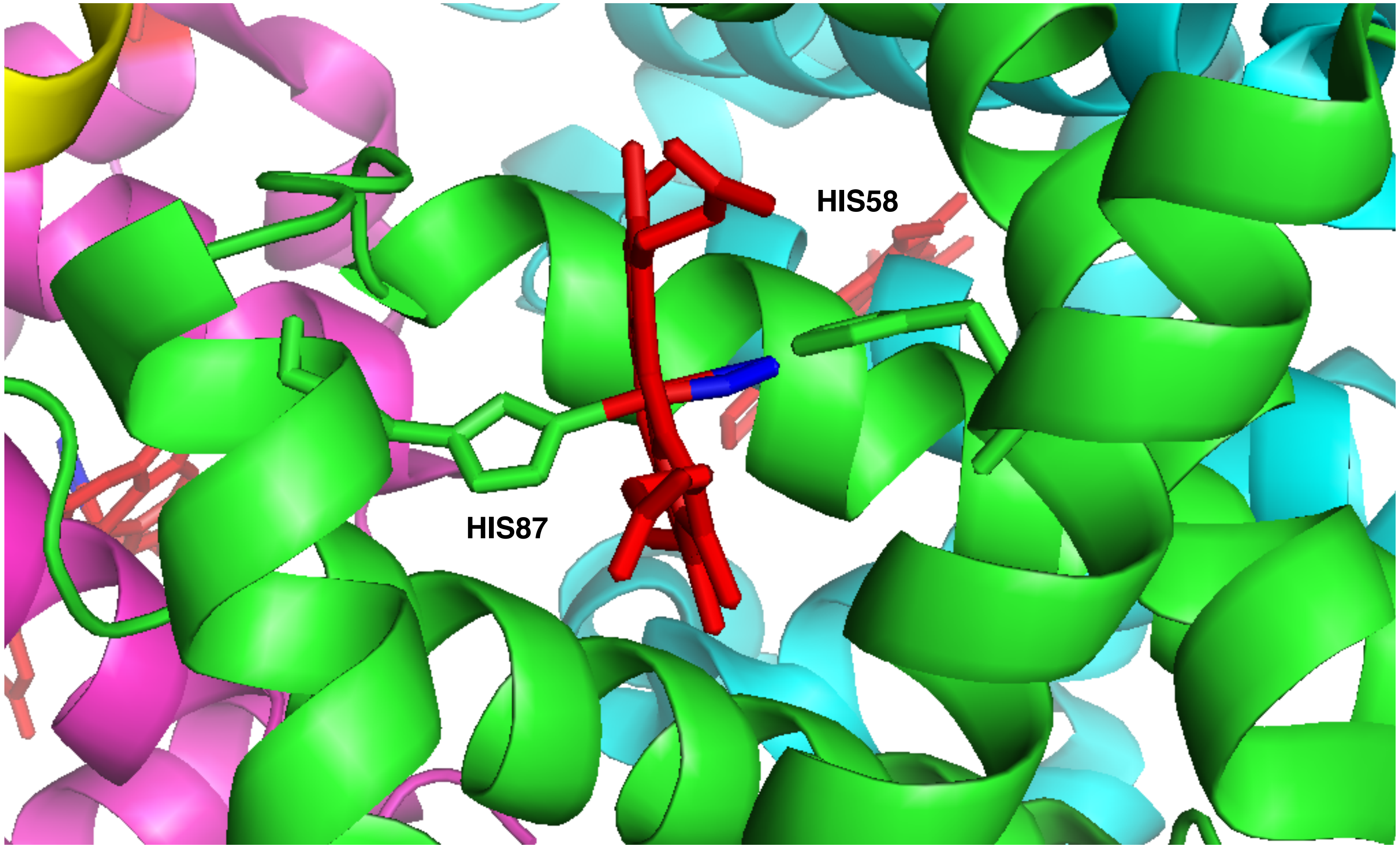
Heme and Oxygen

Anchoring of the heme is facilitated by a histidine nitrogen that binds to the iron. A second histidine is near the bound oxygen.



PDB: 1GZX

Problem 1

Given the PDB structure 1GZX for each monomer:

- Considering a minimum atom distance of 3.5 Å, identify the residues in proximity of the heme and oxygen group.
- Using the same procedure identify the possible interacting residues between monomers.

Suggestions:

- Modify the program for parsing a PDB file including the file ‘HETATM’ that includes the coordinate of the Heme and Oxy groups
- The distance between two group of atoms is the minimum distance between all possible combinations of atoms
- The functions used to calculate the distance between prostetic groups and monomers can be modified to calculate the distances between monomers

Project Goals 1

The main goal of the project is to identify the interactions that are responsible for the formation of the complex and the interaction between the monomers, the heme group and the oxygen bound to the heme group.

Identify the interactions measuring the distances between atoms

- Measure the distances between residues of each monomer, the heme group and oxygen bound to it. Select only atoms below a given threshold (3.5 Å) to identify the interactions with hetero groups (HEM, OXY)

- Measures the distances between residues in different monomers to identify interacting residues. Using a the same threshold indicated above to identify the salt bridges between monomers.

Problem 2

Given the PDB structure 1GZX:

- Generate the dssp file of the whole structure and the substructures and calculate the surface of interaction between monomers.
- Evaluating the differences in relative solvent accessibility, identify the interacting residues between monomers.
- Compare the results obtained for each monomer.

Suggestions:

- Generate the dssp of the single monomers and the four structures excluding one monomer.
- The comparison of the different dssp allows to calculate the surface of interaction that each monomer has with the remaining ones.

Project Goals 2

The main goal of this part of the project is to identify the interactions that are responsible for the formation of the complex considering analyzing the surface of interaction between monomers.

Identify the interactions measuring the loss of solvent accessibility between residues

- Measure the lost of solvent accessibility between the pairs of monomers. Comparing the complex with all the possible trimers obtains removing one monomer at the time. Define the strongest interactions among monomers

- Calculate the lost of solvent accessibility for each residue in the monomers and identify potential interaction hot-spot selecting the hydrophobic residues with high lost of relative solvent accessibility.

Problem 3

Consider the protein-protein interaction network from IntAct database

- Generate the network of human protein-protein interactions supported by direct interaction data (MI:0407)

- Identify the major component of the network and calculate the degree, clustering and betweenness of the Hemoglobin subunits α and β.

- Remove one node at the time and calculate the betweenness of the two nodes.

Suggestions:

- Extract the data from the intact database considering the human taxid 9606

- Replace all the isoforms with the reference protein

Project Goals 3

The main goal of this part of the project consists in the study of the protein-protein interaction of the α and β subunits of the hemoglobin

- Characterize the topology of the network calculating the degree, clustering and betweenness of the Hemoglobin subunits α and β.

- What are the consequences of the removal of the nodes corresponding to the α and β subunits of the Hemoglobin?