# Identification of unknown proteins using structural bioinformatics

## Purpose

Molecular visualization and analysis of 3-D structures of biological molecules.

## Objective

You are required to identify four proteins, (Myoglobin, Cytochorme c, Cytochorme c peroxidase, and Catalase) based on their 3-D structures. Please contact Dr. Dinescu to receive the pdb files (labeled A, B, C, D) of your unknowns.

## Materials and Methods

*Database*: Protein Data Bank (PDB) available at **http://www.rcsb.org/pdb/home/home.do**

*Software*: Swiss PDB Viewer or/and MOE

A short tutorial on how to use these tools will be offered.

## Description

Iron-containing proteins with porphyrin ligands are called heme proteins. While their active sites contain the same Fe-protoporphyrin IX cofactor, these enzymes catalyze different reactions. The geometry of Fe can be five- or six-coordinate (four covalent bonds to the equatorial porphyrin nitrogen atoms and axial ligands above and/or below the equatorial plane), with variable axial ligands among different proteins.
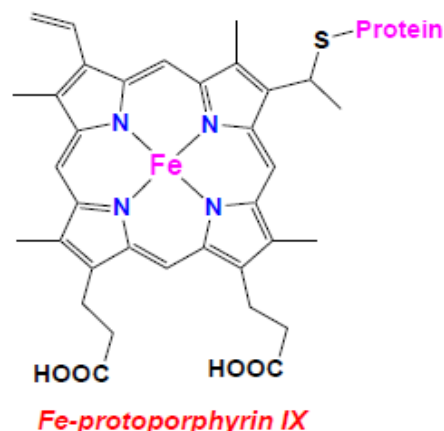
**Fun Stuff:** Molecule of the Month, Protein Data Bank

**Myoglobin -** January 2000 Molecule of the Month
**http://www.rcsb.org/pdb/static.do?p=education_discussion/molecule_of_the_month/pdb1_1.html**

**Cytochrome c -** December 2002 Molecule of the Month
**http://www.rcsb.org/pdb/static.do?p=education_discussion/molecule_of_the_month/pdb36_1.html**

**Catalase -** September 2004 Molecule of the Month
**http://www.rcsb.org/pdb/static.do?p=education_discussion/molecule_of_the_month/pdb57_1.html**

## What to Include in Your Report

Abstract (100 – 250 words)

Introduction: Write a concise overview of the four heme proteins in terms of their functions and reactions (Do not use the "Molecule of the Month" feature from Protein Data Bank).

Methods: Include databases and software used. Describe your work and mention what features you used in your analyses.

Results: For each unknown 3-D structure (A, B, C, and D) list the following parameters:
1) Number (one or two) and identity of axial ligands (sequence number and letter code of the amino acid).
2) The identity of the axial atom(s) (denoted Z below)
3) Protonation state of the axial ligand and its charge
4) Length of Fe-$Z_{axial}$ bonds
5) Length of Fe-$N_{equatorial}$ bonds
6) $Z_{axial}$-Fe-$N_{equatorial}$ bond angles
7) All hydrogen bonds around the axial ligand(s). Give a detailed account on each hydrogen bond
    a. identity of the hydrogen bond partner (sequence number and letter code);
    b. functional group involved in the hydrogen bond and whether is a backbone or side chain group;
    c. hydrogen bond distance (X-H…Y): X-Y distance and H-Y distance in parentheses;
    d. hydrogen bond angle (X-H…Y): X-H-Y bond angle.

In addition, include a clear figure of each active site showing these parameters, and label the amino acids involved in covalent bonding or hydrogen bonding. Discuss the similarities and differences between A, B, C, and D.

**Repeat** the entire procedure using downloaded pdb files from the Protein Data Bank. You are required to decide which structures are to be downloaded. You may download several structures for each protein, but include one structure per protein in your report (do not forget to provide the PDB codes).

Conclusion: Identify the unknown proteins by comparison between your structural parameters. Include sequence alignment pictures to confirm your matches.