**Learning objectives**

1. Analyze the structure of a photosynthetic reaction center using PDB.
2. Identify and evaluate high-probability electron transfer pathways using eMAP.
3. Evaluate cofactors and amino acids that contribute to electron-transfer in proteins.
4. Apply basic concepts from Marcus theory to describe charge separation.
5. Write a report on the background and findings of this activity.

**Resources**

[**Protein Data Bank (PDB) - open access biological macromolecule 3D structural data**](https://www.rcsb.org/)

[**eMAP - identifying and visualizing electron and hole transfer pathways in proteins**](https://emap.bu.edu/)

Bertini, I., Gray, H. B., Stiefel, E. I., Valentine, J. S. (2007). Biological Inorganic Chemistry: Structure and Reactivity. United States: University Science Books.

Lippard, S. J., Berg, J. M. (1994). Principles of bioinorganic chemistry. United States: University Science Books

**Instructions**

Before beginning this activity, students should be familiar with content from Sections X.1.4.2: Multi-Heme Cytochromes, X.2: Electron Transfer through Proteins, and X.3.8: The Photosynthetic Electron-Transfer Chain from Bertini, Gray, Stiefel, and Valentine. Information in Chapter 9: Electron-Transfer Proteins from Lippard and Berg is also useful for this activity.

The crystal structure of the photosynthetic reaction center (PRC) from *Blastochloris viridis*is deposited in the Protein Data Bank (PDB) as 2JBL. You will use this code to find and evaluate structural features of this PRC in PDB and eMAP. As you progress through the instructions below, you should capture screenshots (both PDB and eMAP have built-in screenshot tools) and record observations. With approval from the instructor other photosynthetic reaction centers might be used instead of 2JBL.

You will submit a report containing an introduction to photosynthetic reaction centers, relevant information from Marcus electron transfer theory, and photosynthetic charge separation. After the introduction, you can structure the report however you prefer so long as it includes the following:

* An image of the entire PRC of *Blastochloris viridis*visualizing extracellular, membrane-bound, and intracellular components of the protein assembly.
* An image that clearly visualizes the donor, pigment, intermediate acceptor, and final acceptor (DPIA) system in the PRC.
* Multiple images that clearly convey the optimal electron transfer pathways between nearest neighbors in the PRC DPIA system using the eMAP tool.
* All images should contain a Figure number and caption. You should refer to the Figures in your report as you discuss them.

*Within PDB*

* Find the structure of the PRC of *Blastochloris viridis.*
* Visualize the entire PRC using the "Predict Membrane" feature of the 3D View options.
* Change visualization settings as you prefer. For example, you can hide the polymer to only show cofactors, you can alter cofactor colors to illustrate differences in their structures and the presence of metal ions.
* Your screenshots of DPIA members should include metal ions. eMAP structure renderings do not show metal ions, so use PDB for this purpose.
* One of the acceptor cofactors is the normal quinone (https://en.wikipedia.org/wiki/Vitamin\_K2) but the other is not.
	+ More information about the other cofactor can be found here: https://www.sciencedirect.com/science/article/abs/pii/S0022283607001805

*Within eMAP*

* Find the structure of the PRC of *Blastochloris viridis*using Single Protein Analysis.
* Click through the parameters and ensure appropriate "Non-Protein ET Active Moieties" are selected.
* The general default parameters should be fine but it's good habit to check.
* Select process file to submit the structure to the eMAP electron transfer pathway identification algorithm.
* In the lower left is a graph showing a network of plausible electron transfer pathways. Record observations of the graph. Are electron transfer pathways strictly composed of cofactors? Keep in mind PDB and eMAP will use abbreviated codes for cofactors and amino acids. Hovering over entities in the PDB structure will help you pair a code with a given structure.
* In the lower right is a 3D structure of the protein. At first there will be nothing of interest here. You must specify and submit a source and target to predict the electron transfer pathway(s).
	+ For example, to explore the electron transfer pathway between a donor heme and pigment bacteriochlorophyll, you could specify HEC1335 as the electron source and BCB1275 as the electron target.
	+ After submitting source and target, the graph and structure will update to show a plausible electron transfer pathway by coloring in the electron transfer pathway.
	+ Hover over the cofactors to identify them. Take screenshots of the pathways between D and P, P and I, and I and A.

Your report should contain the following

* **Title page with experiment title and your name**
* **Introduction** to the bacterial PRC, Marcus electron transfer theory, and photosynthetic charge separation.
	+ How do bacteria covert electromagnetic energy into chemical potential energy?
* **Analysis and Discussion**of your screenshots generated in PDB and eMAP.
	+ Discuss whether electron transfer occurs strictly between cofactors or if the protein is directly involved.
	+ If the protein is involved, what amino acid residues are predicted to facilitate electron transfer? What is unique about these amino acid residues the might permit electron transfer?
	+ Find at least one literature reference that addresses the role of the amino acids you identified in photosynthetic reaction centers, and comment on that role in your report.
* **References**
	+ Cite all references using any citation format you wish.