



Department of Chemistry and Biochemistry

**Master of Science
in Biochemistry**

Thesis Defense

Mrs. Jacquelyn M. Miller

B.Sc. Indiana University, South Bend, IN (2014)

Wednesday, November 16, 2016 – 2:00 pm

Byker Auditorium

Department of Chemistry and Biochemistry

“The Role of FixX in Electron Bifurcation”

Graduate Committee

Dr. John Peters (Research Advisor)

Dr. Jennifer DuBois (Chemistry)

Dr. Brian Bothner (Chemistry)

ABSTRACT

Two known methods of physiological energy conservation are substrate level phosphorylation and electron transfer phosphorylation. Recently, electron bifurcation has been established as a third and key mechanism of energy conservation in biological processes. This coupling of endergonic and exergonic reactions allows for utilization of reducing potential to perform energetically expensive physiological reactions. A significant and energetically expensive physiological reaction is nitrogen fixation, which provides a substantial portion of the bioavailable nitrogen that life requires. Electron bifurcation is utilized by the FixABCX system that is up regulated during diazotrophic growth and is suggested to bifurcate electrons from NADH to quinone of the electron transport chain through high potential electron transfer proteins and to nitrogenase through low potential electron transfer proteins. The determination of how cellular mechanisms overcome the energy barriers of high potential electron transfers through electron bifurcation is crucial for our fundamental understanding of energy transfer and energy conservation. The work presented in this thesis aims to progress the present knowledge in this third mechanism of energy conservation and shows support for a protein in the FixABCX complex, FixX, as the low potential electron acceptor in the complex. Numerous organisms were investigated as potential model systems for FixABCX with varying degrees of success. The genome of the organism, *Roseiflexus castenholzii*, contains both the nitrogenase and *fixABCX* genes and has successfully been used to obtain FixX. This protein shows homology to ferredoxin, a physiological reductant of the nitrogenase Fe protein in some organisms. EPR spectroscopy and sequence analysis suggests FixX contains 2 [4Fe-4S] clusters, while a potentiometric titration shows the clusters to have highly negative mid-point potentials. The preliminary evidence supports FixX of the FixABCX system to be a low potential electron transfer protein.

BIOGRAPHICAL NOTES

Academic Preparation:

2009-2014 Indiana University, South Bend, IN (May 2014)
Bachelor of Science in Biochemistry
Senior Seminar: Pea Seeding Copper Amine Oxidase
Advisor: Dr. Gretchen Anderson

Graduate Studies

Field of Study: Biochemistry

Courses

2014 X-Ray Crystallography, Virology
2015 Proteins, Advanced Genetics, Microbial Physiology
2016 Mass Spectrometry

Teaching and Outreach Activities

2015 General Chemistry Lab TA, Montana State University (Jan.-May)
2014 Biochemistry Lab TA, Montana State University (Aug.-Dec.)
2014 Science Alive Youth Outreach, River Park Library
2012 Science Fair Judge, St. Anthony de Padua Catholic Church and School
2012 Science Fair Judge, St. Monica Catholic School
2012 Guest Lecturer General Chemistry, Indiana University

Awards

2014 MSU Meritorious Award
2013 IUSB Academic Centers for Excellence Outstanding Tutor Award
2013 IUSB Student Services Employee of Excellence Award
2012 IUSB Student Services Employee of Excellence Award