Understanding the reaction mechanism of Cytochrome c oxidase in *Rhodobacter sphaeroides*

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Understanding the reaction mechanism of Cytochrome c oxidase in *Rhodobacter sphaeroides*

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Introduction

A Typical Animal Cell

Mitochondria

**Mitochondria**

- **Glucose → Glycolysis**
  - Pyruvate → Lactate
  - Acetyl-CoA → NADH

- **Krebs Cycle**
  - NADH
  - Electron transport system

- **ETs – Electron Transport System**
  - ATP

**Oxidation**

\[ \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{O}_2 \rightarrow 6\text{CO}_2 + 6\text{H}_2\text{O} + \text{energy} \]

**Reduction**

**Glycolysis**

**ETS**

**Krebs Cycle**

**ETS**

**ATP**

**ETS – Electron Transport System**
Introduction

Electron Transport System (ETS)
Introduction

Electron Transport System

Complex I

Complex III
(Cytochrome $bc_1$)

Complex VI
(Cytochrome $c$ oxidase)

$$2\text{NADH} + 2\text{H}^+ + \text{O}_2 \rightarrow 2\text{H}_2\text{O} + 2\text{NAD}^+$$
Cytochrome c oxidase

Terminal membrane protein.

It catalyzes the reduction of oxygen to water.

It utilizes the free energy of the reduction reaction for pumping the proton across the inner-mitochondrial membrane.

\[
4\text{Cyt } c^{\text{red}} + 8\text{H}^+_{(\text{in})} + \text{O}_2 \rightarrow 4\text{Cyt } c^{\text{ox}} + 2\text{H}_2\text{O} + 4\text{H}^+_{(\text{out})}
\]
Understanding the reaction mechanism of Cytochrome c oxidase

*Rhodobacter sphaeroides*

**Description of the project**

- Understanding the reaction mechanism of Cytochrome c oxidase
- *Rhodobacter sphaeroides*
Goals:

- How the $O_2$ reduction coupled with $H^+$ pumping?
- What are the important residues involved in this mechanism?
- Along what path the protons are transferred to the binuclear center?
- What are the pathways for protons which are pumped across the membrane?
Description of the project

Proposed catalytic cycle for Cytochrome c oxidase

- DFT calculations to calculate the pKₐ values of the active sites in O and E states.
- Determination of protonation of the protein in the O and E states using electrostatic calculations.
- Similar calculations for R, Pₘ, and F states.
- Redox calculations for tyrosine.
- Electrostatic calculation for Pₘ state; where does the radical go!!!
Present Study

$pK_a$ and redox calculations on cross-linked Tyrosine-Histidine in Cytochrome c oxidase
Present Study

$pK_a$ calculations on cross-linked Tyrosine-Histidine

*Experimental $pK_a$ values from Donk et al. (J. Am. Chem. Soc. 2000, 122, 2403–2404)*
Summary

- The catalytic cycle of Cytochrome c oxidase is still unclear.

- DFT (Density Functional Theory) and electrostatic calculations to determine the pK_a values and redox potentials of the protein in the different intermediate states.

- Applying the kinetic Monte Carlo method to understand the proton pumping in Cytochrome c oxidase.
Thanks for your Attention !!!

Prof. G. Matthias Ullmann
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