Photoinduced electron transfer in cytochrome bc₁: Kinetics of ubiquinone transfer from the Qₒ site to the Qᵢ site, and evidence for communication between the monomers in the dimer

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The electron transfer reactions within wild-type Rhodobacter sphaeroides cytochrome bc₁ (cyt bc₁) were studied using a ruthenium dimer to rapidly photo oxidize cyt c₁. It was found that when cyt bₜ was initially reduced before the reaction, photooxidation of cyt c₁ led to bifurcated reduction of both the iron-sulfur protein and cyt bₜ by QH₂ in the Qₒ site, followed by re-oxidation of two equivalents of cyt bₕ and cyt bₜ. It was proposed that the newly formed ubiquinone diffused through the hydrophobic cavity linking the Qₜ site of the reactive monomer A to the Qᵢ site of the other monomer B, leading to oxidation of cyt bₜ in monomer B followed by oxidation of cyt bₕ in monomer A by cross-monomer electron transfer. Addition of one equivalent of the Qᵢ site inhibitor antimycin to the cyt bc₁ dimer had very little effect on any of the electron transfer reactions, while addition of a second equivalent completely inhibited re-oxidation of cyt bₜ and cyt bₜ. It was also found that addition of one equivalent of the Qₒ site inhibitor stigmatellin to the cyt bc₁ dimer completely inhibited all electron transfer reactions in both monomers of the dimer. These experiments are consistent with a half-of-the-sites mechanism in which only one monomer of the dimer is active at a time, implying monomer-monomer interactions. The rapid electron transfer reaction from the ISP to cyt c₁ was found to be greatly decreased by viscosity, indicating a multi-step diffusional mechanism as the iron-sulfur protein rotates from the b state to the c₁ state.

Biography
Francis Millett received his BS in Chemistry from the University of Wisconsin in 1965, his PhD in Chemical Physics from Columbia University in 1970, and was an NIH Postdoctoral Fellow at California Institute of Technology from 1970-1972. He joined the faculty of the University of Arkansas in 1972, and is now a Distinguished Professor. He developed, together with Bill Durham, the ruthenium photoreduction method which made it possible to measure the kinetics of key steps in electron transfer during mitochondrial oxidative phosphorylation. He has directed collaborative, multidisciplinary research which combines rapid kinetics methods, site-directed mutagenesis, X-ray crystallography, and NMR to investigate protein structure-function relationships.

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