

Picosecond fluorescence decay of tryptophan in bovine cytochrome-c oxidase

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Detailed fluorescence studies on bovine heart cytochrome oxidase (CcO) has been carried out in lauryl maltoside solution. Steady state fluorescence of the tryptophan residues of the enzyme showed that the fluorphores are embedded deep inside the hydrophobic protein cavity. Time resolved studies of tryptophan fluorescence of native and heat treated CcO have been carried out for the first time using synchronously pulsed picosecond dye laser by single photon counting technique. Decay of the tryptophan fluorescence has been fitted using discrete four exponential model. Amplitude distribution of lifetimes also showed four distinct regions in the maximum entropy calculation of the decay profiles. Results of the time resolved studies have been analyzed using a model of distribution of the tryptophan residues in four distinct domains with respect to the heme centres. Fluorescence energy transfer of the tryptophan residues to the heme centres have been used to determine average distances of different domains from the heme centres in CcO. A comparison of the results of the native and heat treated CcO has been used to identify any structural changes in the enzyme due to heat treatment.

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