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Electron Paramagnetic Resonance Study of the Complex Dynamics of the Spin Label Attached to Cytochrome C.

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Measurements of an EPR signal of a nitroxide spin label attached to a protein allow for exploring the motion and structural changes of the macromolecule at the level of the back-bone fold. X-band EPR spectra of the spin label attached to the iso-1-cytochrome c at position 102 (recorded for temperatures between 4–40°C) contain two components with different mobility. They were analyzed using "the microscopic order –macroscopic disorder" model (MOMD). The spectra of the spin labeled protein, either dissolved in a sucrose solution or bound to the CM Cellulose gel, have been compared. Experimental lineshapes have been successfully fitted to the theoretical model. Two components of the spectra are characterized by different values of rotational diffusion rates and ordering potential coefficients. Their values increase with temperature, but the relative fraction of either component in the spectrum is independent of temperature. The mean correlation times characterizing the faster component resolved in the EPR spectrum are different for these two kinds of samples. Analysis of the prototype system: "cytochrome c electrostatically bound to the gel" prepares the way for study the interaction of this protein with cytochrome bc1, one of the three major respiratory en-zyme complexes.