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Authors

GRATTON, E
SILVA, N
ROSATO, N
[et al.](#)

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Enrico Gratton, Norberto Silva Jr, Nicola Rosato, Alessandro Finazzi-Agrò, and Giampiero Mei.

Molten globule in superoxide dismutase.

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Abstract

The enzyme, human superoxide dismutase is a homodimer containing a single solvent-exposed tryptophan residue per subunit. This protein denatures as the concentration of guanidine hydrochloride is increased to about 6M. The midpoint of the transition is at about 3.5M guanidine hydrochloride for the holo protein. The denaturation process is accompanied by a dissociation of the dimeric native protein into monomers. The monomer-dimer equilibrium can be independently followed by changing the total protein concentration of the solution. We have studied the fluorescence and circular dichroism (CD) properties of the monomer and we have shown that in the monomer state the protein has the typical characteristics of a molten globule. In the molten globule state, the CD signal in the 220 nm region is identical to the CD signature of the native protein. By contrast, in the aromatic region around -270 nm the CD signal in the monomer state is lost. The fluorescence properties show that in the molten globule state the fluorescence decay is very heterogeneous, indicating the existence of a disorganized tertiary structure. Also, the decay of the emission anisotropy is relatively fast, as compared with the native protein, indicating a fluid-like environment for the surrounding of the tryptophan residue. Supported by NIH RR03155 and the Italian CNR.