

Cloning, Purification and Biophysical Analysis of Manganese Superoxide Dismutase from the Filamentous Fungus *Trichoderma reesei*

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Fungi are capable of metabolizing aromatic compounds by oxidative reactions that may involve superoxide dismutase (SOD), but there is no work described for the filamentous fungus *Trichoderma reesei* to date. To explore this gap in the literature we cultivated *T. reesei* cells in the presence of benzonitrile. Total RNA was isolated for cDNA synthesis and random clone analysis allowed identifying a putative manganese superoxide dismutase. The length of TrSOD-Mn coding sequence is 639 bp and it encodes a protein of 212 aminoacids (approximately 23.2 kD). The primary sequence of the protein was aligned with other fungi orthologs and it presented strongly similarity (above 67 %). TrSOD-Mn was expressed in *E. coli* and purified using affinity chromatography for further studies using circular dichroism (CD). The CD spectrum of native TrSOD-Mn in the far UV region suggests the presence of helical structure content, indicated by a maximum at 195 nm and a broad minimum between 210 and 220 nm. Deconvolution analysis showed 52% of helical structure and a minor content of beta structures (23%), turns (13%) and unordered forms (14%). CD spectra also indicated that TrSOD-Mn preserves its secondary structure when submitted to temperature variation at 30 °C and 50 °C for one hour of incubation. At temperature 80 °C, the enzyme maintains its fold up to 40 min of incubation. Although our results provide preliminary data on TrSOD-Mn they help to target future research for enzymatic and biophysical characterization of this enzyme.

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