Cloning, Expression, and Purification of a Cu/Zn Superoxide Dismutase from *Jatropha curcas*

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We report cDNA cloning, expression, purification, and characterization of a novel Cu/Zn superoxide dismutase (SOD) from *Jatropha curcas* leaves. The full-length cDNA of this SOD contained a 496-bp open-reading frame (ORF) encoding 162 amino acid residues. The recombinant plasmid containing the SOD coding sequence was introduced into *Escherichia coli*, and the SOD was expressed as a fusion protein. The recombinant SOD was purified from a high-density fed-batch culture using a combination of immobilized metal ion affinity chromatography (IMAC) and Sephadex G25 desalting chromatography. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) analysis indicated that the recombinant SOD was a monomeric protein with a molecular mass of approximately 16.4 kDa. Isoelectric focusing showed that this SOD was a basic protein with p*I* values of 7.04, 7.33, 8.62, and 8.77. The activity of the SOD was stable at 70 °C for 10 min, and in a broad pH range from 4 to 9. The presence of urea (up to 8 M), guanidinium chloride (up to 6 M), and 2-mercaptoethanol (up to 8 mM) had little effect on the activity. The activity decreased gradually with increasing concentrations of imidazole, hydrogen peroxide, and ethylenediaminetetraacetic acid (EDTA). Atomic absorption spectrometry showed the presence of 0.239 copper and 0.258 zinc atoms, respectively, in the SOD polypeptide.

Key words: Superoxide Dismutase, *Jatropha curcas*