Molecular cloning, prokaryotic expression and functional characterization of superoxide dismutase gene from *Siniperca chuatsi*

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ABSTRACT Superoxide dismutase is an important antioxidant enzyme in organisms. Copper/Zinc superoxide dismutase, designated as ScCu/Zn-SOD was identified from mandarin fish *Siniperca chuatsi*. The full-length cDNA of ScCu/Zn-SOD was 804 nucleotides with an open-reading frame of 465 bp encoding 154 amino acids. The deduced amino sequence of ScCu/Zn-SOD had no signal peptide, which included two conservative signatures (^{45}GFHVHVFGDN^{55} and ^{139}GNAGGRLACGVI^{150}) of Cu/Zn-SODs family. ScCu/Zn-SOD shared high degree of identity (60.13-92.21%) with intracellular Cu/Zn-SODs from other species. ScCu/Zn-SOD mRNA was widely expressed in all tissues of *S. chuatsi*, including muscle, gill, liver and kidney, etc., the muscle and gill were higher level of expression, but lower level of expression were in head kidney and spleen, relatively.

The recombinant expression plasmid (pET-30a+ScCu/Zn-SOD) was constructed by inserting coding region of ScCu/Zn-SOD gene into pET-30a expression vector, and transformed it into *E. coli* BL12 strains (DE3). The natural soluble recombinant proteins of ScCu/Zn-SOD by inducing with 0.5 mM IPTG, 0.5 mM $CuSO_4$ and 0.1 mM $ZnCl_2$ at 20 ℃. The concentration of recombinant protein after purification was 0.14 mg/mL, and the Cu/Zn-SOD enzymatic activity was 108.5 U/mg. The further research of recombinant proteins enzyme activity indicated that it was stable at 25-60 ℃ and pH 5.0-9.0, and could resistant to 5% SDS.

Key words: *Siniperca chuatsi*; Superoxide dismutase; Gene cloning; Prokaryotic expression; Recombinant protein.

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