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A N I N T E R N A T I O N A L J O U R N A L

Molecular And 3D-Structural Characterization Of Fructose-1,6-Bisphosphate Aldolase Derived From *Metroxylon Sagu*

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ABSTRACT

Fructose-1,6-bisphosphate aldolase (FBAld) is an enzyme that catalyzes the cleavage of D-fructose-1,6-phosphate (FBP) to D-glyceraldehyde-3-phosphate (G3P) and dihydroxyacetone phosphate (DHAP), and plays vital role in glycolysis and gluconeogenesis. However, molecular characterization and functional roles of FBAld remain unknown in sago palm. Here we report a modified CTAB-RNA extraction method was developed for the isolation of good quality RNA (RIN>8) from sago leaves and the isolation of FBAld cDNA from sago palm. The isolated sago FBAld (msFBAld) cDNA has total length of 1288 bp with an open reading frame of 1020 bp and a predicted to encode for a protein of 340 amino acid resides. The predicted protein shared a high degree of homology with Class-I FBAld from other plants. Meanwhile, the msFBAld gene spanned 2322 bp and consisted of five exons. Conserved domain search identified fifteen catalytically important amino acids at the active site and phylogenetic tree revealed localization of msFBAld in the chloroplast. A molecular 3D-structure of msFBAld was generated by homology modeling and a Ramachandran plot with 86.7% of the residues in the core region, 13.4% in the allowed region with no residues in the disallowed region. The modeled structure is a homotetramer containing an α/β-TIM-barrel at the center. Superimposition of the model with Class-I aldolases identified a catalytic dyad, Lys209-Glu167, which could be involved in the Schiff's base formation and aldol condensation. Apart from that, overproduction of the recombinant msFBAld in Escherichia coli resulted in increased tolerance towards salinity.

Key words: *Metroxylon sagu*; Fructose-1,6-bisphospate aldolase; 3D-structure; stress; Schiff's base

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