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α -Amylase from *Pseudoalteromonas* sp. MY-3: Cloning, Expression and Enzymatic Properties

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The gene encoding an extracellular α -amylase from *Pseudoalteromonas* sp. MY-3, a marine bacterium, was cloned, sequenced and expressed in *Escherichia coli*. Nucleotide sequence analysis showed an open reading frame of 2007 bp which could encode a polypeptide comprised of 669 amino acids. The protein has a predicted PI of 5.15. The nucleotide sequence and the amino acids sequence of this amylase gene showed high degree of similarity with α -amylase preproprotein gene from *Pseudoalteromonas haloplanktis*, 86% identities, and α -amylase of *Pseudoalteromonas atlantica*, 69% identities, respectively. The molecular weight of the gene was detected about 75 kDa by SDS-PAGE.

Key words: α -Amylase, *Pseudoalteromonas* sp., cloning, expression

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Molecular Cloning of Soybean *MMP25* Gene Encoding Matrix Metalloproteinase Protein Induced by Abiotic Stresses

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Matrix metalloproteinase proteins (MMPs) are involved in remodeling of plant extracellular matrix in association with plant growth, development, and possibly defense processes. A novel soybean (*Glycine max*) metalloproteinase gene, *MMP25* was identified. The complete cDNA sequence of *MMP25* comprised of 1,443 bp with an open reading frame of 1,179 bp which encodes 43.2 kDa polypeptide consisting of 393 amino acid residues. The nascent *MMP25* polypeptide contained N-terminal signal peptide with a central hydrophobic core between amino acids Asp₂₉ and Ser₃₀, and predicted cleavage site between amino acids Asp₁₅₃ and Val₁₅₄. The deduced *MMP25* polypeptide is a pre-pro-enzyme that has all of the hallmark motif characteristic of matrix metalloproteinases. To confirm the expression of the *MMP25* gene at the transcriptional level, northern blot analysis was also carried out using the mRNA prepared from developing soybean cotyledons and the soybean leaves exposed to various stresses and hormone. The expression of *MMP25* was induced by LT (5 °C), NaCl, wounding stresses and ABA. *MMP25* protein expressed in *E. coli* cells showed protease activity in zymography assay. Further *in vivo* function of *MMP25* was investigated in *MMP25* overexpressing *Arabidopsis* plants.

Key words: Soybean, metalloproteinase, stress, gene expression