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Molecular and biochemical analysis of soybean *Gm8244* induced by low temperature

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L-asparaginase mediates the conversion of asparagine into aspartate and ammonia, and plays an important role in nitrogen metabolism in plants. It also has a role in biosynthesis of amino acids and nodulation process in legumes. *Gm8244* clone has high homologies with L-asparaginase gene. In this study, the functional analysis of *Gm8244* gene is performed. The function of *Gm8244* gene was analyzed through *E. coli* transformation. The recombinant proteins purified with three-step purification schemes; fractionation, immobilized metal ion affinity chromatography, and histidine affinity chromatography. Recombinant *Gm8244* protein was expressed in *E. coli* cells. L-asparaginase activity of *Gm8244* was assayed by measuring ammonia amount released from asparagine. *E. coli* cells expressing *Gm8244* function showed an increased L-asparaginase activity. The protein of antisense pET-*Gm8244* was largely increased the amount of asparagine compared with sense pET-*Gm8244*.

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Differential expression of soybean *Gm2256* gene by low temperature

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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 noble soybean (*Glycine max*) metalloprotein gene, *Gm2256* was identified. The complete cDNA sequence of *Gm2256* comprised 1,443 bp with a initiation codon at position 102 and a termination codon at position 1,259. The *Gm2256* sequence has an open reading frame of 1,179 nucleotides which encodes 43.2 kDa polypeptide consisting of 393 amino acid residues. The nascent *Gm2256* 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 *Gm2256* polypeptide is a pre-pro-enzyme that has all of the hallmark motif characteristics of matrix metalloproteinases. To confirm the expression of the *Gm2256* gene at the transcriptional level, Northern blot analysis was also carried out using the mRNA prepared from the soybean leaves exposed to various stresses and hormone. The expression of *Gm2256* is induced by LT(5°C) at 24 hrs and ABA at 6 hrs. The highest induction of *Gm2256* occurs by NaCl-treatment for 6 hrs.