

Public Abstract

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Title:MOLECULAR CLONING AND CHARACTERIZATION OF REGULATORY ENZYMES IN THREONINE BIOSYNTHETIC PATHWAY FROM SOYBEAN

Soybeans are a good source of protein and oil. Despite being a very good source of protein, soybeans nutritional value is limited by low proportion of sulfur amino acids methionine and cysteine demanding supplementation of soy-based animal feed with synthetic amino acids. The cost involved aside, other essential amino acids, called second-tier amino acids, are also becoming limiting in animal feed due to this practice. Threonine is one of those second-tier amino acids and improving its proportion in soybean seeds can improve the nutritional value of soy-based animal feed. The manipulation of regulatory enzymes in the biosynthesis pathway of this amino acid is one method used to increase the proportion of free threonine in soybean seeds. One prerequisite for such an approach is the isolation and characterization of the genes encoding the key enzymes. The goal of this project was to isolate and characterize genes coding two regulatory enzymes- homoserine dehydrogenase (HSDH) and threonine synthase (TS)- in threonine biosynthesis pathway

The genes coding both HSDH and TS have been isolated and named gmhsdh and gmts, respectively. The gmhsdh seems to code a mono-functional, cytosolic HSDH and is interrupted by 11 introns. The gmts encodes a chloroplast localized TS and this gene has no introns. The cDNA encoding HSDH was also isolated and expressed in E.coli to verify the functionality of the predicted coding region of HSDH and used to raise antibodies. Multiple sequence analysis of the HSDH amino acid sequence leads to a proposition that this protein is feedback insensitive to threonine. Verification of this fact could make this a very good candidate for achieving a favorable proportion of not only threonine but also methionine and isoleucine in the free amino acid pool. The promoter region of gmts also was isolated and sequenced, which could reveal novel mechanisms in the regulation of plant TS.