

Why does 3-hydroxy-3-methylglutaryl coenzyme A lyase not interfere with cytoplasmic mevalonate biosynthesis in plants?

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Little is known on the function of 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) lyase (HMGL, EC 4.1.3.4) in plants, other than it might participate in leucine degradation and the mevalonate shunt.¹ The enzyme had been partially purified from a heavy membrane fraction isolated from 4-d-old etiolated radish seedlings, but proved too unstable to study in great detail its molecular and kinetic properties.² After the total sequence of the Arabidopsis genome was available, it seemed more appropriate to isolate a putative cDNA clone from this closely related plant, which was then further characterized. Arabidopsis expresses two differentially spliced mRNAs coding for two putative isoforms, having either a molecular mass of 51 kDa (HMGL51) or of 46 kDa (HMGL46). HMGL46 contains an unusual N-terminal extension of 100 amino acids with no targeting properties, which seems to be specific to plant enzymes. In contrast, the intact HMGL51 contains a mitochondrial leader peptide. Transient expression of GFP fusion proteins in tobacco cells shows that the enzyme can either be directed to peroxisomes because of the presence of a C-terminal peroxisomal retention motif and/or to mitochondria, depending on the expressed form and the position of the additional 100 amino acid peptide within the protein. This latter sequence bears no similarity with any other protein sequence in data banks. When heterologously expressed in *Escherichia coli*, only a truncated (35 kDa) protein (HMGL35) led to an active enzyme, catalyzing the formation of acetyl-CoA and acetoacetate from HMG-CoA, and its kinetic properties have been examined. This truncated form displays a high degree of sequence similarity with vertebrate and bacterial HMGL enzymes, which suggests an important metabolic role. HMGL35 without any targeting motif was shown to be cytotoxic when induced in transformed Arabidopsis plants. In its high molecular mass and thus inactive form, the cytoplasmic mevalonate pathway might be protected from interference with HMGL during the protein's transit to the target organelles.

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