

Thiamine biosynthesis in *Arabidopsis thaliana* reveals an intricate pattern of evolution and involvement of distinct subcellular compartments

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Thiamine or vitamin B1 is an essential nutrient required by all living cells. Organisms capable of synthesising thiamine de novo include plants, fungi, and many microorganisms, where it is formed by the coupling of two independently synthesised molecules, thiazole phosphate and pyrimidine pyrophosphate. However, the understanding of the pathways involving thiamine biosynthesis (TBS) in plants is still incipient. Using yeast two-hybrid techniques we have identified two *Arabidopsis thaliana* sulphur transferases with the potential to interact with AtTH1, the only plant protein previously known to be involved in the synthesis of thiazole. Assays using the thiamine antagonist oxythiamine show that the *Arabidopsis At1g02880* gene product is functionally equivalent to *Saccharomyces cerevisiae* thiamine pyrophosphokinase, and transient expression assays reveal a cytosolic localisation. The AtHMPPK/TMPPase however, was localised to both the plastids and mitochondria in onion epidermis transient expression assays. Phylogenetic analysis revealed some of the evolutionary origins of the TBS proteins in plants showing that the pathway in plants can be thought of as a mosaic and is in fact made up of steps with differing evolutionary origins.

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