Functional Analysis of *EMB* Genes Using Epitope-Tagged Proteins

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An important goal of genomics research is to acquire a comprehensive understanding of protein interactions inside the living cell. Advances in protein purification techniques and mass spectrometry have significantly accelerated the detection and analysis of multiprotein complexes, particularly in model eukaryotes. In our SeedGenes Project (www.seedgenes.org), we have confirmed the identities of more than 30 *EMB* genes predicted to encode proteins with unknown or uncertain functions. These represent a valuable subset of *Arabidopsis* genes for tag-based affinity purification methods because the identification of molecular partners may help to reveal their underlying functions. We describe here the molecular complementation of mutant embryos with histidine-tagged (6xHis) and tandem affinity purification-tagged (TAP) proteins. We demonstrate that 6xHis- and TAP-tagged EMB1629 protein can restore normal function in the absence of competing native protein, enabling the recovery of viable homozygous knockouts. The TAP-tagged protein has been detected by western blot analysis and work is underway to isolate potential interacting partners. We also present the results of molecular complementation experiments involving other *EMB* genes predicted to encode proteins with unknown functions. The epitope tag in some cases appears to interfere with normal protein function because the mutant phenotype is only partially rescued. We conclude that complementation of knockout mutants with TAP-tagged proteins, when followed by recovery and identification of interacting protein partners, represents an effective strategy for understanding the biological functions of many essential genes in *Arabidopsis*.

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