



**CALIFORNIA STATE SCIENCE FAIR
2006 PROJECT SUMMARY**

Name(s) Vinay Tripuraneni	Project Number S0426
Project Title Characterization of the Arabidopsis thaliana plsp1-1 and plsp1-2 Lines: Toc75 Suggests a Novel Protein Targeting Pathway	
<p style="text-align: center;">Abstract</p> <p>Objectives/Goals Toc75 forms the only known protein translocation channel in the chloroplast outer envelope membrane, and is thus crucial to plastid biogenesis. Toc75 is an unusual transmembrane protein in that it has 2 transit peptides; most membrane targeted proteins have none. A type 1 signal peptidase, Plsp1, is proposed to cleave the second transit peptide.</p> <p>Methods/Materials When PLSP1 is disrupted/knocked out with a tDNA insertion in the fourth intron, plants of the line, plsp1-1, exhibit a pale phenotype with underdeveloped chloroplasts. However, when the same gene is disrupted with a tDNA insertion in the first exon, plants of the line, plsp1-2, exhibit a phenotype similar to the wild type plants.</p> <p>Results The tDNA insertion portion of plsp1-2 was sequenced, and found to be immediately after the first methionine of the Plsp1 sequence. Because the beginning of Plsp1 has more than one methionine, interrupting the first methionine may not cause a knockout of PLSP1. This was examined by reverse-transcriptase mediated PCR, which revealed that the plsp1-2 plants homozygous for the tDNA insertion are transcribing PLSP1. Immunoblotting and chloroplast import in Pisum sativum of Toc75 suggests that Plsp1 is the protease responsible for cleaving the second transit peptide of Toc75.</p> <p>Conclusions/Discussion This evidence reveals a novel protein targeting mechanism where Toc75 demonstrates a completely new model for protein targeting pathways in all organisms. This new pathway has ramifications beyond chloroplast protein import; it may provide a model for new and more sensitive tests for genetic illnesses, give further insight into membrane chemistry and drug delivery, and may also be a step towards a cure for peroxisomal and mitochondrial disorders.</p>	
Summary Statement Proteins make up nearly everything in an organism. A new way to get proteins where they are supposed to go (protein targeting) has been discovered!	
Help Received Used lab equipment at California State University, Fresno under supervision of Dr. Calderon-Urrea. Also used lab equipment at University of California, Davis under supervision of Dr. Inoue	