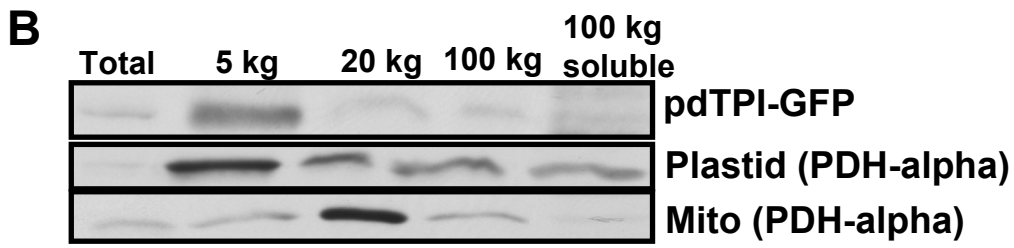
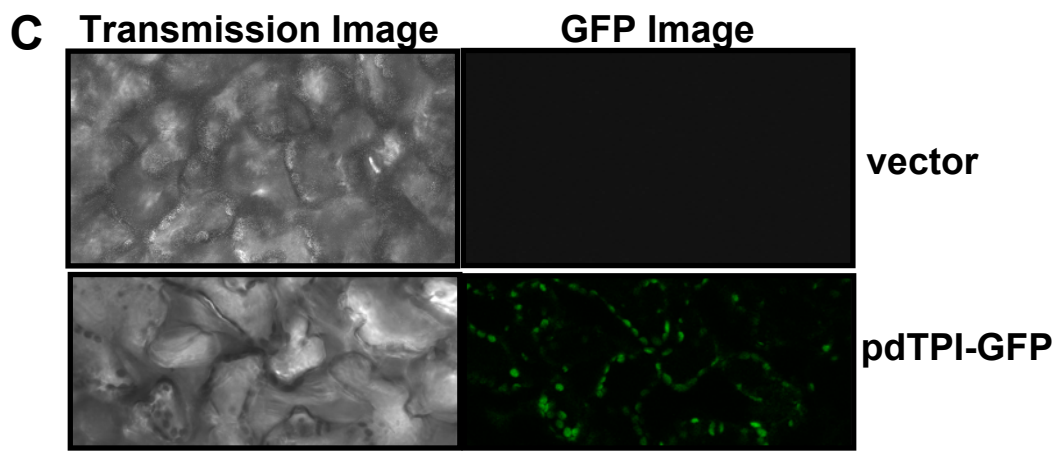
A

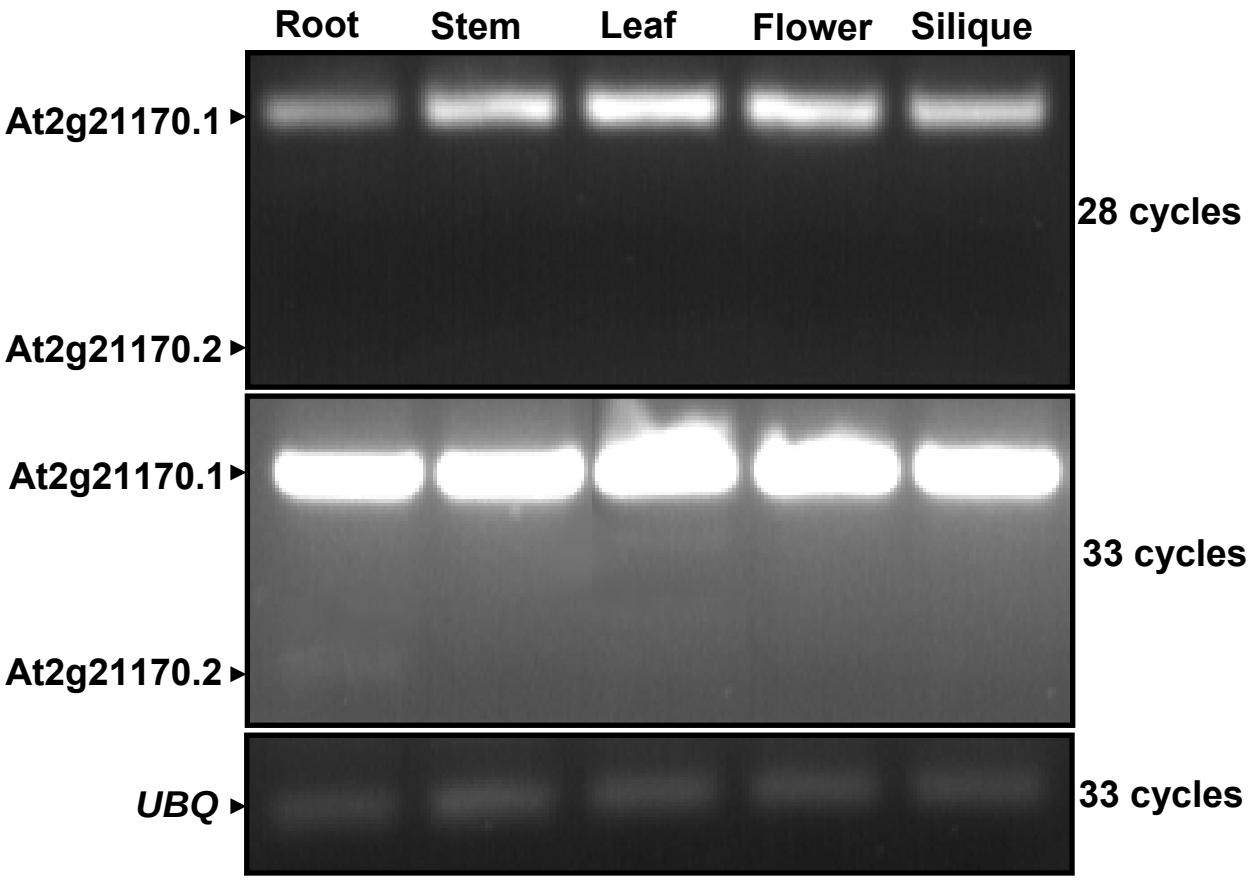
```
gi602589          ----------------------------MGRKFFVGGNWKC-------------------- 13
gi38112661        ----------------------------MGRTFFVGGNWKC-------------------- 13
AtCytoTPI         ----------------------------MARKFFVGGNWKC-------------------- 13
gi11124571        ----------------------------MGRKFFVGGNWKC-------------------- 13
gi407524          ----------------------------MGRKFFVGGNWKC-------------------- 13
gi1785947         ----------------------------MGRKFFVGGNWKC-------------------- 13
gi169820          ----------------------------MGRKFFVGGNWKC-------------------- 13
gi77540215        ----------------------------MGRKFFVGGNWKC-------------------- 13
gi57283984        ----------------------------MGRKFFVGGNWKC-------------------- 13
gi84626306        ----------------------------MARKFFVGGNWKC-------------------- 13
gi115440976       ----------------------------MAARKFFVGGNWKC-------------------- 14
gi340520          ----------------------------MGRKFFVGGNWKC-------------------- 13
AtPdTPI           MAATSLTAPPSFSGLRRISPKLDAAAVSSHQSFFHRVNSSTRLVSSSSSSHRSPRGVVAM 60
                                                :.**   * .

gi602589          -----NGTAEEVKKILATLNAADVPSQDVVEVVVSPPYVFLPLVKNELRPDFHVAAQNCW 68
gi38112661        -----NGTSEEIKKIVATLNAGQVPSQDVVEVVVSPPYVFLPLVKNELRSDFHVAAQNCW 68
AtCytoTPI         -----NGTAEEVKKIVNTLNEAQVPSQDVVEVVVSPPYVFLPLVKSTLRSDFFVAAQNCW 68
gi11124571        -----NGTVEQVESIVNTLNAGQIASTDVVEVVVSPPYVFLPTVKGKLRPEIQVAAQNCW 68
gi407524          -----NGTVSQVETIVNTLNAGQIASPDVVEVVVSPPYVFLPTVKDKLRPEIQVAAQNCW 68
gi1785947         -----NGTVEQVEAIVQTLNAGQIVSPDVVEVVVSPPYVFLPIVKAKLRPEIQVAAQNCW 68
gi169820          -----NGTTDQVDKIVKILNEGQIASTDVVEVVVSPPYVFLPVVKSQLRPEIQVAAQNCW 68
gi77540215        -----NGTTEEVKKIVTTLNEAKVPGEDVVEVVVSPPFVFLPFVKSLLRPDFHVSAQNCW 68
gi57283984        -----NGTTEEVKKIVTTLNEAKVPGEDVVEVVVSPPFVFIPTVKTLLRPDFHVAAQNCW 68
gi84626306        -----NGTVAEVNKIVKLLNEADVPSEDVVEVVISPPFVFLPQVKAVLRSDFAVAAQNAW 68
gi115440976       -----NGTGEDVKKIVTVLNEAEVPSEDVVEVVVSPPFVFLPQVKGLLRPDFSVAAQNCW 69
gi340520          -----NGTSEEVKKIVTLLNEAEVPSEDVVEVVVSPPYVFLPFVKNLLRADFHVAAQNCW 68
AtPdTPI           AGSGKNGTKDSIAKLISDLNSATLEAD--VDVVVSPPFVYIDQVKSSLTDRIDISGQNSW 118
                       ***  .:  ::  ** . : .   *:**:***:*::  **  *   : ::.**.*
```

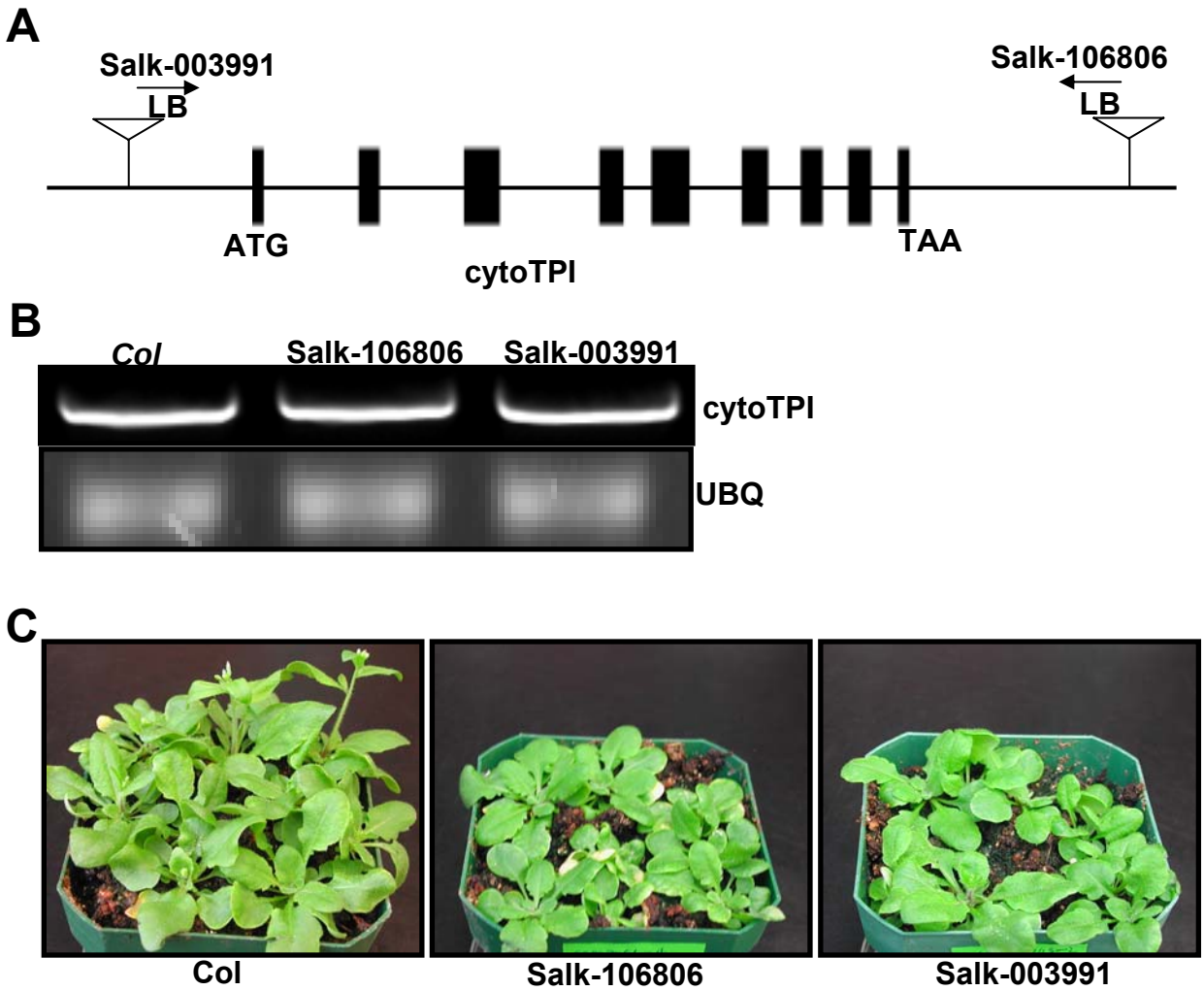




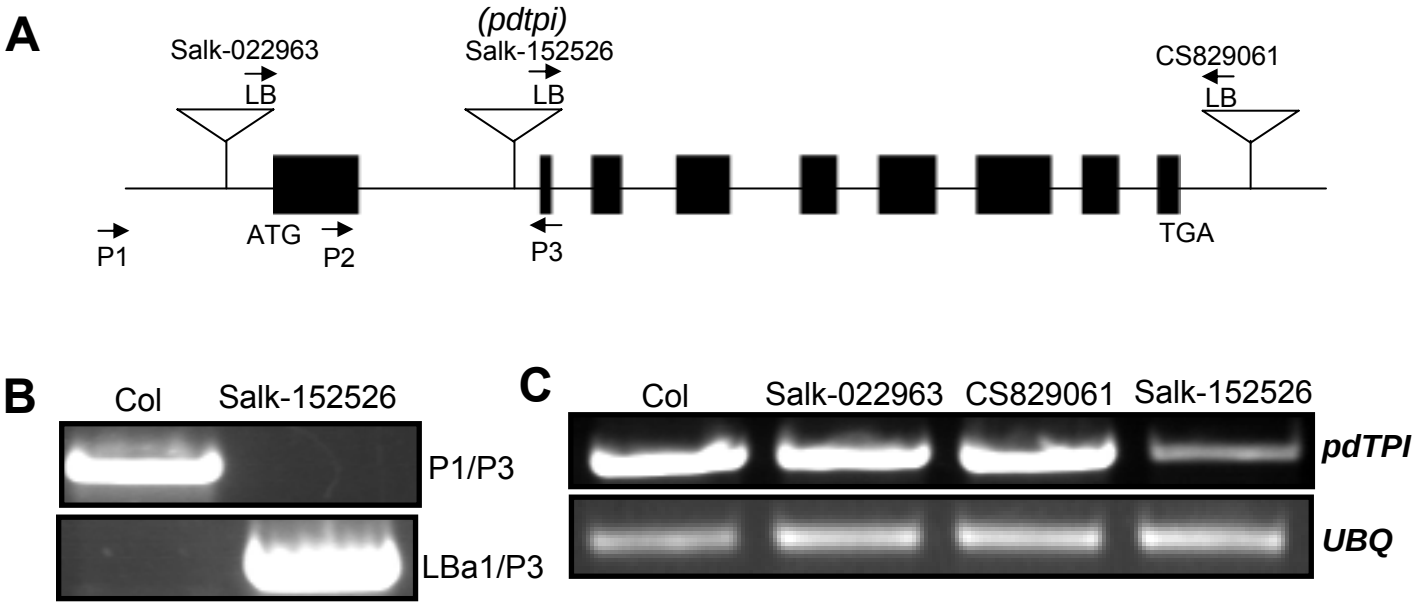
Supplemental Figure 1. pdTPI protein localizes to the chloroplast. **(A)** Alignment of *pdTPI* amino acid sequence with proteins from Arabidopsis (At cytoTPI), Petunia (gi602589), *Solanum chacoense* (gi38112661), *Coptis japonica* (gi340520), *Oryza sativa* (gi169820; gi115440976), *Triticum aestivum* (gi11124571), *Glycine max* (gi77540215), *Phaseolus vulgaris* (gi57283984), *Secale cereale* (gi407524), *Hordeum, vulgare* (gi1785947), *Pteris vittata* (gi84626306). **(B)** Subcellular localization of the pdTPI:GFP fusion protein in leaf cells. Transgenic Arabidopsis leaf protein expressing pdTPI-GFP was fractionated and detected by immunoblotting. Plastid PDH α-subunit was used to determine plastid enrichment; antibody to the mitochondrial PDH α-subunit was used to assay mitochondria enrichment. **(C)** Leaves from the transgenic pdTPI-GFP lines were observed by confocal microscopy. Transgenic lines transformed with vector only were used as a control.



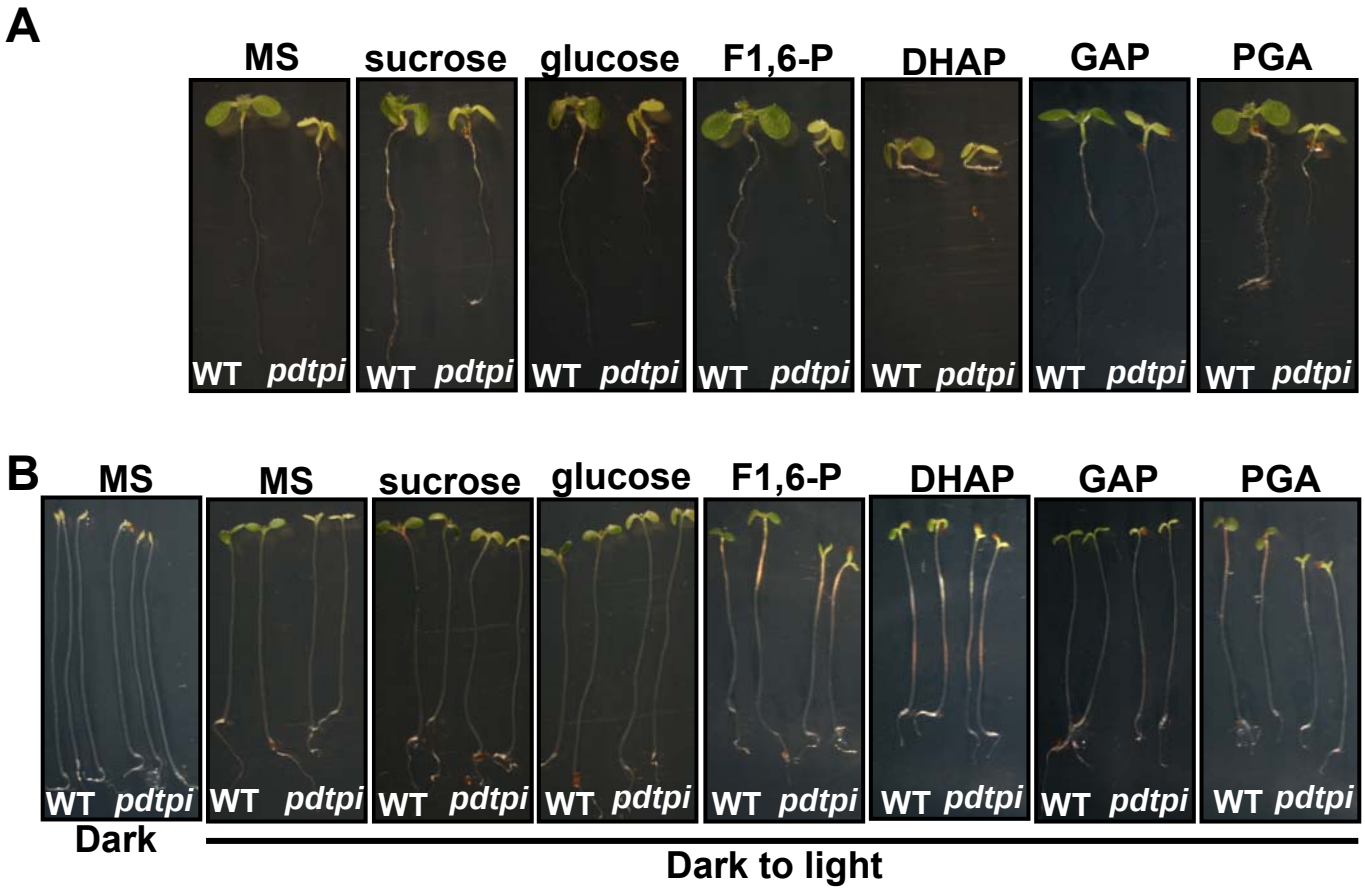
Supplemental Figure 2. Expression of alternatively spliced *pdTPI* transcripts.
Tissues were harvested from *Col* wild-type. RT-PCR was applied to detect transcript abundance. Primers S21170F and S21170R were used to amplify 122 bp and 95 bp fragments from splice variant 1 and splice variant 2, respectively. *UBQ* was used as loading control.



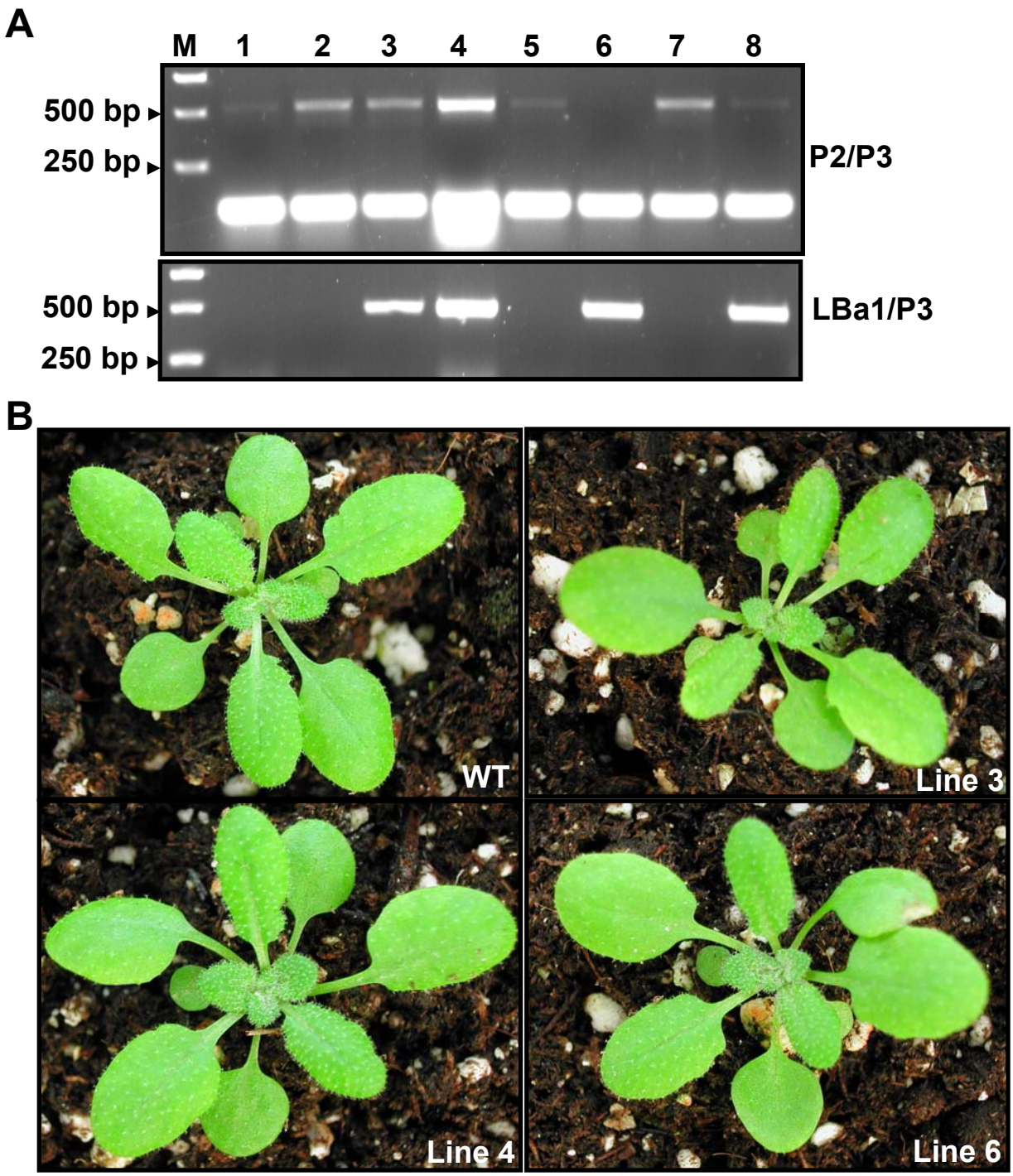
Supplemental Figure 3. CytoTPI mutant identification.
(A) Schematic representation of T-DNA insertion positions in At3g55440 gene. LB, T-DNA left border.
(B) *cytoTPI* gene expression was not affected in Salk-106806 and Salk-003991 T-DNA insertion lines.
(C) Salk-106806 and Salk-003991 appeared similar to wild type when grown for 7 weeks under long-day conditions.



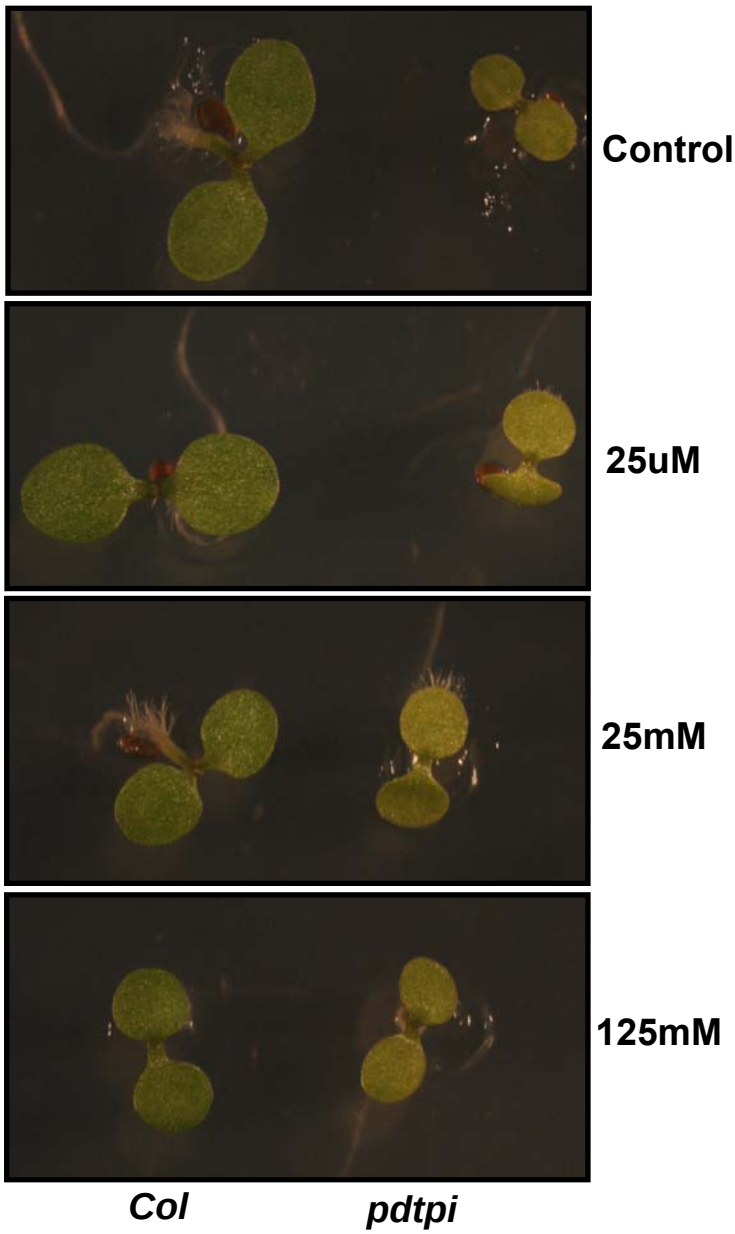
Supplemental Figure 4. Characterization of *pdtpi* mutant.
(A) Schematic representation of T-DNA insertion positions in At2g21170 gene. LB, T-DNA left border. The primer locations and directions were indicated here.
(B) T-DNA insertion in Salk-152526 was verified by PCR by combining T-DNA primer and plant gene-specific primers.
(C) RT-PCR results indicated that T-DNA insertion in Salk-152526 line reduced gene expression. *pdTPI* gene expression in Salk-022963 and CS829061, however, was not affected. *UBQ* was used as a control.



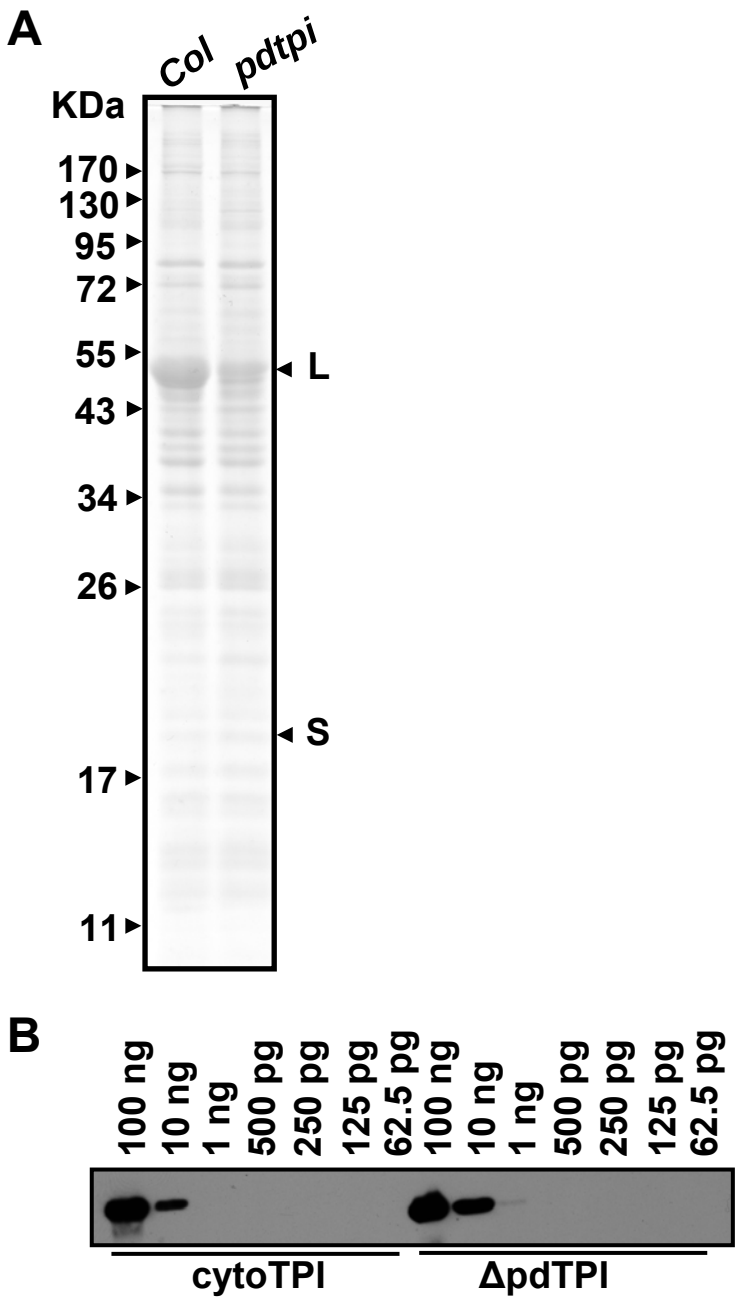
Supplemental Figure 5. Metabolite augmentation does not rescue the *pdtpi* mutant phenotype.
(A) Plants germinated on MS agar plates with the addition of 1 mM of sucrose, glucose, fructose-1, 6-bisphosphate (F-1,6-P), dihydrohyacetone phosphate (DHAP), DL-glyceraldehyde 3-phosphate (GAP) and D(-)3-phosphoglyceric acid (PGA) for 5 d.
(B) Plants were first germinated on regular MS plates under darkness for 5 d, and then were transferred onto MS plates enriched with glycolytic intermediates and grown for an additional 2 d under continuous white light.



Supplemental Figure 6. Complementation of *pdtpi* mutant by transgenic expression of *pdTPI* cDNA.
(A) PCR genotyping of individual transgenic T1 lines.
(B) Wild type and complementation lines grew under long-day condition for 20 days; the T2 generation of rescued lines 3, line 4 and line 6 are shown here.



Supplemental Figure 7. *pdtpi* mutant can not be rescued by the application of myo-inositiol.
Wild type and *pdtpi* mutant were germinated on myo-inositol enriched MS plates for 5 d under continuous white light.



Supplemental Figure 8. Rubisco large subunit is down-regulated in the *pdtpi* mutant.
(A) Total protein from 5-d old seedlings was isolated and separated by SDS-PAGE. L: Rubisco large subunit, S: Rubisco small subunit. Anti-cytoTPI polyclonal antibody cross-reacts with pdTPI protein.
(B) Different amounts of purified, recombinant cytoTPI and ΔpdTPI protein were separated by SDS-PAGE, transferred to nitrocellulose membrane and probed with anti-cytoTPI antibody.