Supplementary Table S1

Primers for making probes for RNA gel blot hybridizations

Arabidopsis	Forward Primer	Reverse Primer
16S	ATGGATACTAGGCGCTGTGC	GTAATACGACTCACTATAGGGACCTTCCTCCGG CTTATCAC
23S	CCTAGATGGCGAGAGTCCAG	GTAATACGACTCACTATAGGGAAGACTCGCTTTC GCTACG
petL	ATTTCAATTGAAACTTAGGG	GTAATACGACTCACTATAGGGAAATTTGGTAATT AACACGG
rpl14	ACAGCGGGGCTAGAGAATTG	GTAATACGACTCACTATAGGGTGGGATCGCCCC AAAAACTC
rpl16	AGACAAATAGAAGCAGGG	GTAATACGACTCACTATAGGGCTATATTTTCGGG TACAC
rps8	GGAAACATCGCGAAAACAACC	GTAATACGACTCACTATAGGGCGAGCTTCTCGG TCTGTCATT
Maize	Forward Primer	Reverse Primer
rpl14	TATACCGTCGTCGCCTTTGA	CCCCTGGTTTTTCTTTCTGG
rps8	GGGCAAGGACACTATTGCTG	CATTCCGCCCAAAACCTTAG
rps3	CATTCCTTTTGGTTCGCACA	GCGAATTGTTTGGAGGGGTA
petL	GGGGACTCATGTTCCGTCTGA	CCCACGAATCTCAATGACCA
ycf4	CAATTCCTTGTGCGGGATATC	GCAAGAAATAGGCCAATTCG
psaI	TCGAATAGATAGAAATAGTAA	CTAGACAATCTTATTTTTCTGC
psaI-ycf4 A oligo probe	GCTATAGTCGGCTACGCATATGCATGCATTTATGCTC GTAGCTTATATCT	
psaI-ycf4 B oligo probe	ACTTTCTATTCAAAAAAAATTAGTGGATCCACTTAAAA AAAAAAACTC	
psaI-ycf4 C oligo probe	CTAGTACTCCTGTGAAATAATTGGTTAGATACATTGA CTTTC	
psaI-ycf4 D oligo probe	GATTTTTCACTTTACTTTTTTTGATTCTGAAATAGCCTT CAGTAGCTAGTAC	

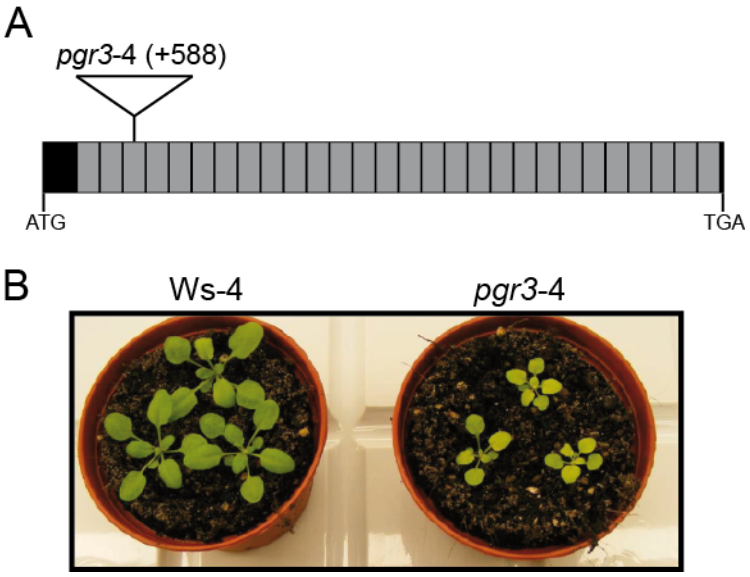
Primers for cloning, genotyping and cRT-PCR in maize

tk121pgr35'	TCCACTGGGCTCTTGGTAGC
tk781pgr33'	CAAGAACCCGAATGCAGATG
PGR3 BamHI-For	CACGGATCCGACGAACGGCGGGTCGTGGGCACGGACAGCG
PGR3-1807R	GGAGGATAGTACACAGTGTGGC
PGR3-1254F	GCAGCTGCTCGAAGAAATGACACG
PGR3-HindII-Rev	CACCTCGAGCTACAATTGATTTGGAAGTTGC
tkrpl14_207for2	ATTCAAAGGCGACGACGGTA
rpl16PE	CTAACAGTCACACACTAAGCAT

Primers for 3'RACE, Arabidopsis

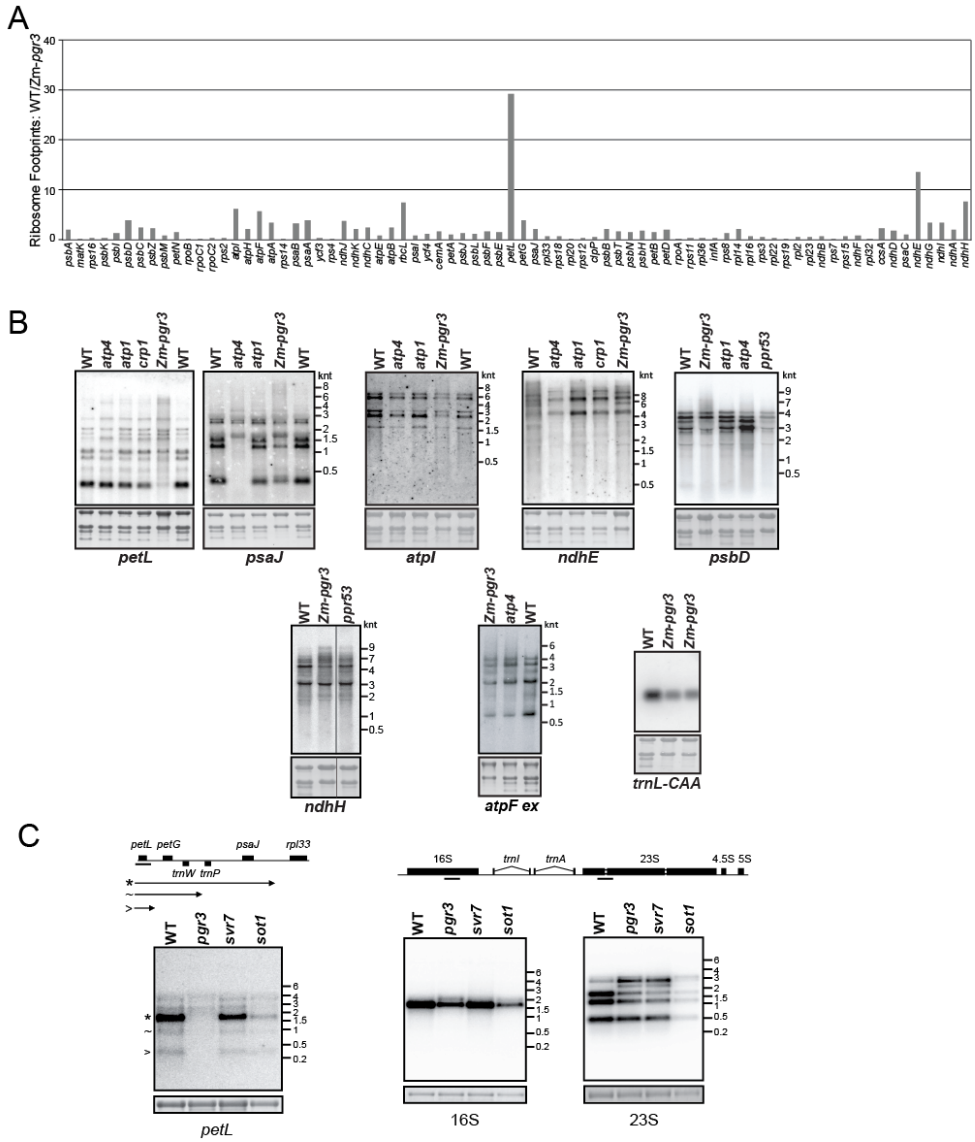
Adapter for 3' RACE	[Phos]GCNNNNTCGTATGCCGTCTTCTGCTTGC[23ddC]
PCR primer	CAAGCAGAAGACGGCATACG
rpl14.rp	ACAGCGGGGCTAGAGAATTG
RT primer	CAAGCAGAAGACGGCATA

Supplementary Figure S1



Supplementary Figure S1. Overview of the *pgr3-4* allele.
(A) Diagram of *PGR3* (At4g31850) showing the position of the T-DNA insertion in the *pgr3-4* allele (FLAG_086D06). The 28 PPR repeats of PGR3 are indicated by grey rectangles. The nucleotide position of the T-DNA insertion with respect to the start codon is indicated in parentheses.
(B) Phenotype of wild type (ecotype Ws-4) and *pgr3-4* plants. Seeds were sown in soil and grown for 23-days under 16h light (120µmol x m-2 x sec -1)/ 8h dark cycles at 23˚C.

Supplementary Figure S2



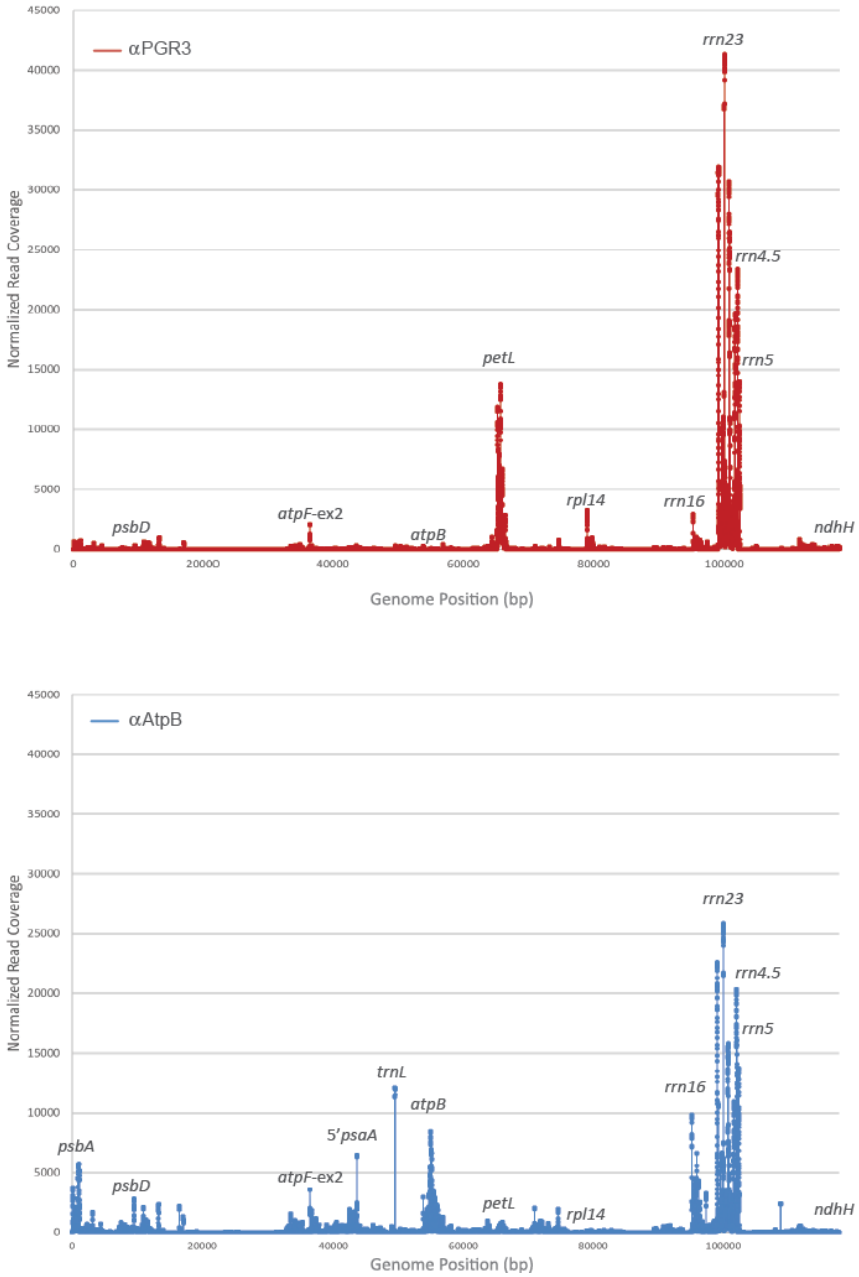
Supplementary Figure S2. Comparison of chloroplast ribosome footprints in Zm-*pgr3* and wild-type plants and additional RNA gel blot analyses of *pgr3* mutants.

(A) The data for Zm-*pgr3* are the same as those shown in Figure 2B, but are plotted as a ratio with respect to the signal from wild-type siblings. Although *ndhE* expression was reduced in comparison with the wild-type, *ndhE* was expressed similarly in the Zm-*pgr3* and *cps1* mutant (Figure 2B) suggesting that this is a secondary effect of Zm-*pgr3's* chloroplast ribosome deficiency.

(B) Additional RNA gel blot hybridizations of Zm-*pgr3* mutants. The methylene-blue stained blots are shown below to illustrate rRNA abundance as a loading control. The *petL* data was reported previously (21). Various other non-photosynthetic maize mutants were included as controls: *crp1* (56), *atp4* (37), *atp1* (54), and *ppr53* (41).

(C) RNA gel blot hybridization analyses of the Arabidopsis *pgr3* mutant. The methylene-blue stained blots are shown below to illustrate rRNA abundance as a loading control. The *svr7* (35) and *sot1* (42) mutants (orthologous to maize *atp4* and *ppr53,* respectively), were included as controls.

Supplementary Figure S3



Supplementary Figure S3. Genome-wide view of Zm-PGR3 RIP-seq data. Read counts were normalized to the total number of reads mapping to the chloroplast genome, and the normalized values mapping to each 10-nt window across the chloroplast genome is plotted. An immunoprecipitation with an antibody to AtpB served as a negative control. The AtpB antibody immunoprecipitated *atpB* mRNA, presumably via interaction with the nascent peptide exposed from translating ribosomes. The chloroplast rRNAs may derive from ribosomes associated with immunoprecipitated mRNAs, from direct interaction of PGR3 with rRNA sequences, and/or from contamination.