

DGCR14 –At3g07790

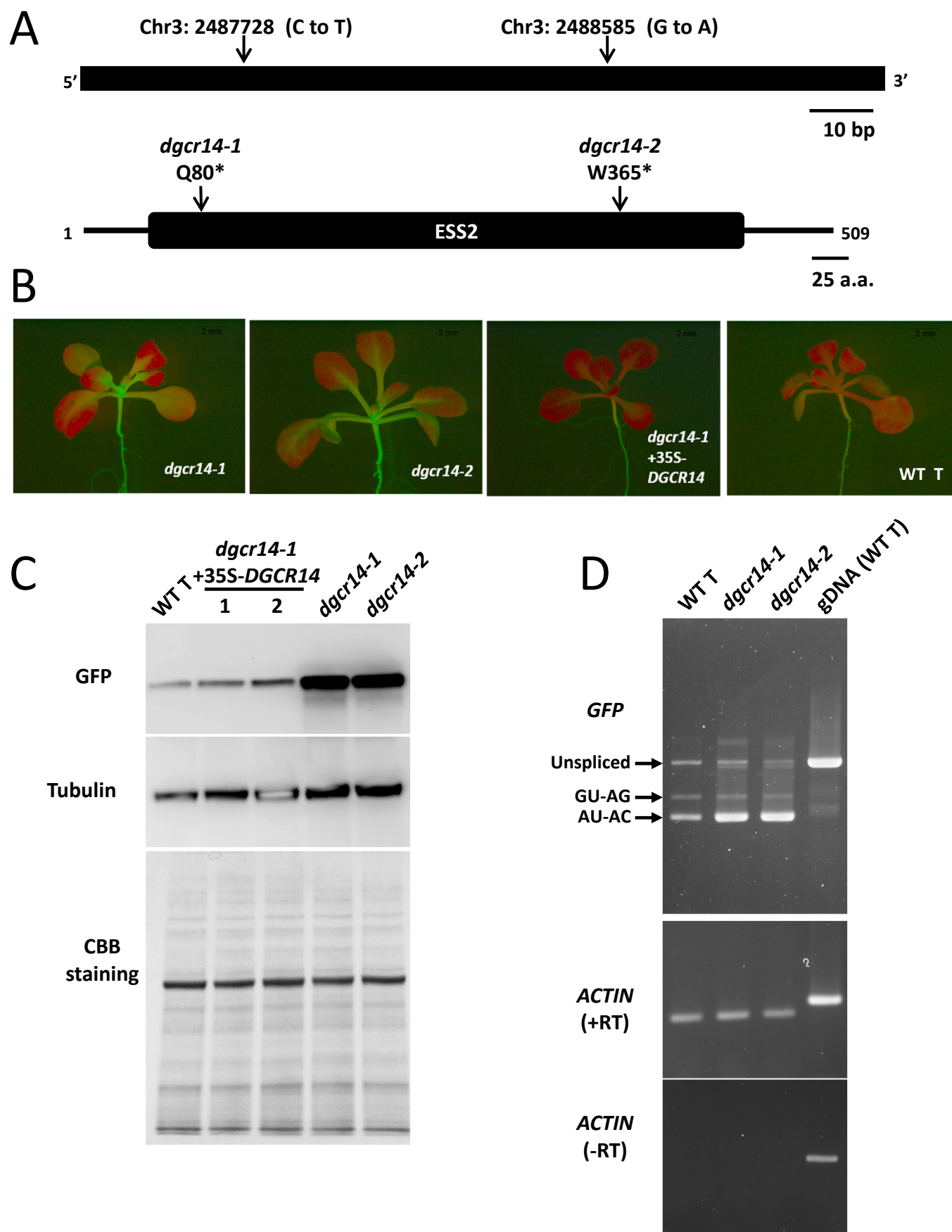


Figure S2: DGCR14

- A. Top: Intron-exon structure of the *DGRC14* gene (At3g07790) and positions of nucleotide changes in the *dgcr14* mutants. Bottom: Position of the ESS2 domain of the DGCR14 protein (509 amino acids) and positions of premature termination codons (asterisks) resulting from the *dgcr14-1* and *dgcr14-2* mutations.
- B. GFP fluorescence in seedlings with the indicated genotypes. The two *dgcr14* mutants are hyper-GFP relative to the WT T line. A complemented *dgcr14-1* mutant, which shows a return to more WT T levels of GFP, is labeled with '+35S-DGCR14' (35S denotes the 35S promoter of cauliflower mosaic virus)
- C. Western blot showing increased levels of GFP protein, consistent with a hyper-GFP phenotype, in the two *dgcr14* mutants relative to the WT T line. The GFP levels in complemented plants (*dgcr14-1*+*DGCR14*, two complemented lines shown) return to approximately the wild-type level. Antibodies to tubulin were used to probe the blot to control for loading levels, which were also demonstrated by coomassie brilliant blue (CBB) staining of the gel.
- D. Semi-quantitative RT-PCR indicating levels of the three *GFP* splice variants in the two *dgcr14* mutants. The level of the translatable AU-AC transcript increases in the mutants, suggesting increased splicing efficiency of the non-canonical splice sites in *GFP* pre-mRNA. Actin, with and without reverse transcriptase (RT), was used as a constitutively expressed control. gDNA, genomic DNA control