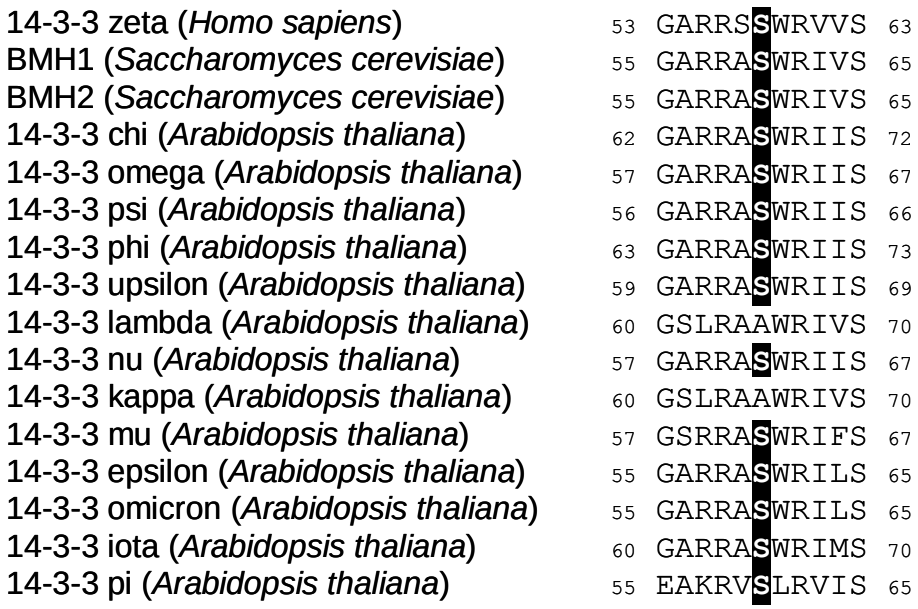

Figure S1. A serine phosphorylation site is conserved among various members of the 14-3-3 family

The region flanking the residue Ser58 in human 14-3-3 zeta (ζ) was aligned with that of all 14-3-3 isoforms from *Saccharomyces cerevisiae* and *A. thaliana*. The conserved serine residue is highlighted in black.

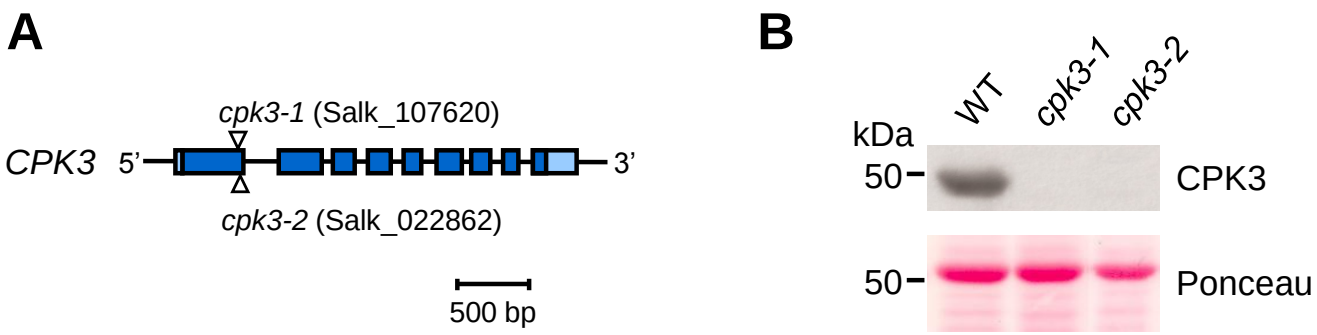

Figure S2. Two different T-DNA insertion lines for CPK3, *cpk3-1* and *cpk3-2*, are null mutants at the protein level

(A) Schematic representation of the *Arabidopsis CPK3* gene and its alleles *cpk3-1* and *cpk3-2*. Boxes indicate exons and dark blue represents the coding region. The positions of T-DNA insertions are marked by triangles.
(B) Protein extracts from leaves of *Arabidopsis* wild type (WT) or mutant plants (*cpk3-1*, *cpk3-2*) were analysed by immunoblotting with an anti-CPK3 antibody and Ponceau staining. The results presented are representative of 3 separate experiments.

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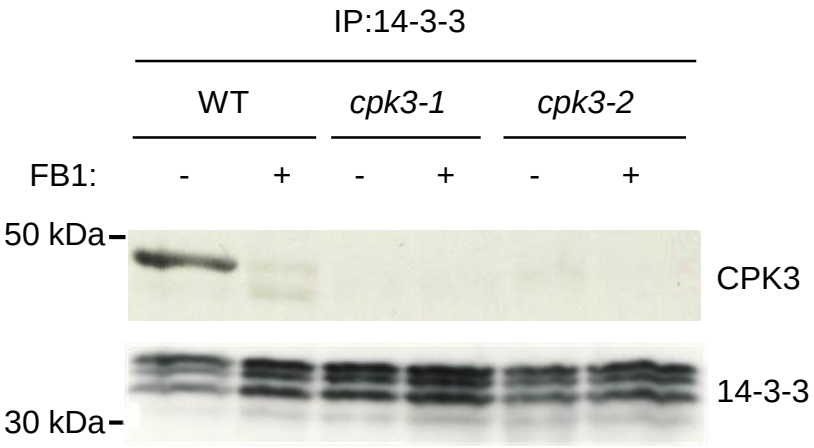

Figure S3. CPK3 is not associated with 14-3-3s in *cpk3* mutant plants

Arabidopsis leaves of wild type (WT) or mutant plants (*cpk3-1*, *cpk3-2*) were treated +/- 10 μM FB1 for 48 hr. 14-3-3 immunoprecipitates were immunoblotted with the indicated antibodies. The results presented are representative of 3 separate experiments.

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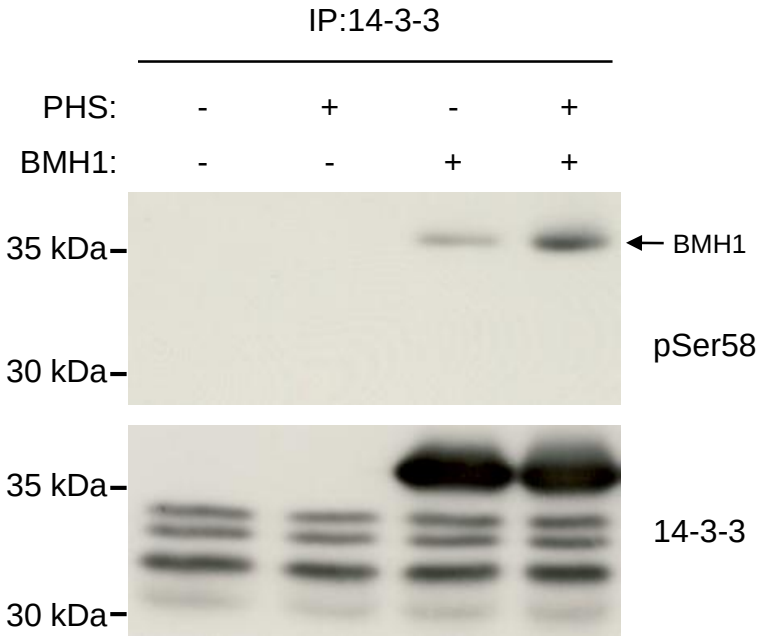

Figure S4. A 14-3-3-associated kinase phosphorylates 14-3-3 Ser58 site of the yeast 14-3-3 BMH1 in a PHS-dependent manner

Proteins from untreated *Arabidopsis* cells were immunoprecipitated with an anti-14-3-3 protein antibody. Immunoprecipitates were used as kinase sources in nonradioactive *in vitro* phosphorylation assays performed in the presence or not (+/-) of recombinant BMH1 and +/- 25 μM PHS. Proteins from these assays were blotted with the indicated antibodies. The results presented are representative of 3 separate experiments. The upper panel showing detection of pSer58 corresponds to a shorter exposure than the blot in Figure 4A, left panel.

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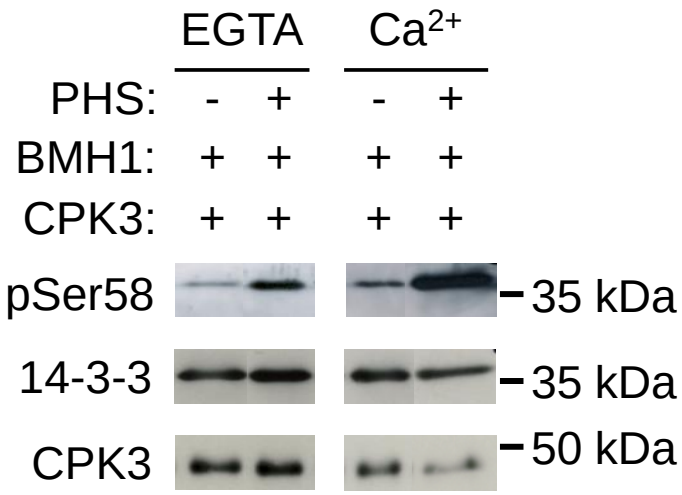

Figure S5. CPK3 phosphorylates 14-3-3 Ser58 site of the yeast 14-3-3 BMH1 in a PHS- and calcium-dependent manner

Kinase assays were performed with recombinant *Arabidopsis* CPK3 and BMH1 (used as the substrate) in the presence or absence of 25 μM PHS, and in the presence (Ca^{2+}) or absence (EGTA) of Ca^{2+}. Proteins were analysed by immunoblotting with the indicated antibodies. The results presented are representative of 3 separate experiments. The right upper panel showing detection of pSer58 corresponds to a longer exposure than the blot in Figure 4C.