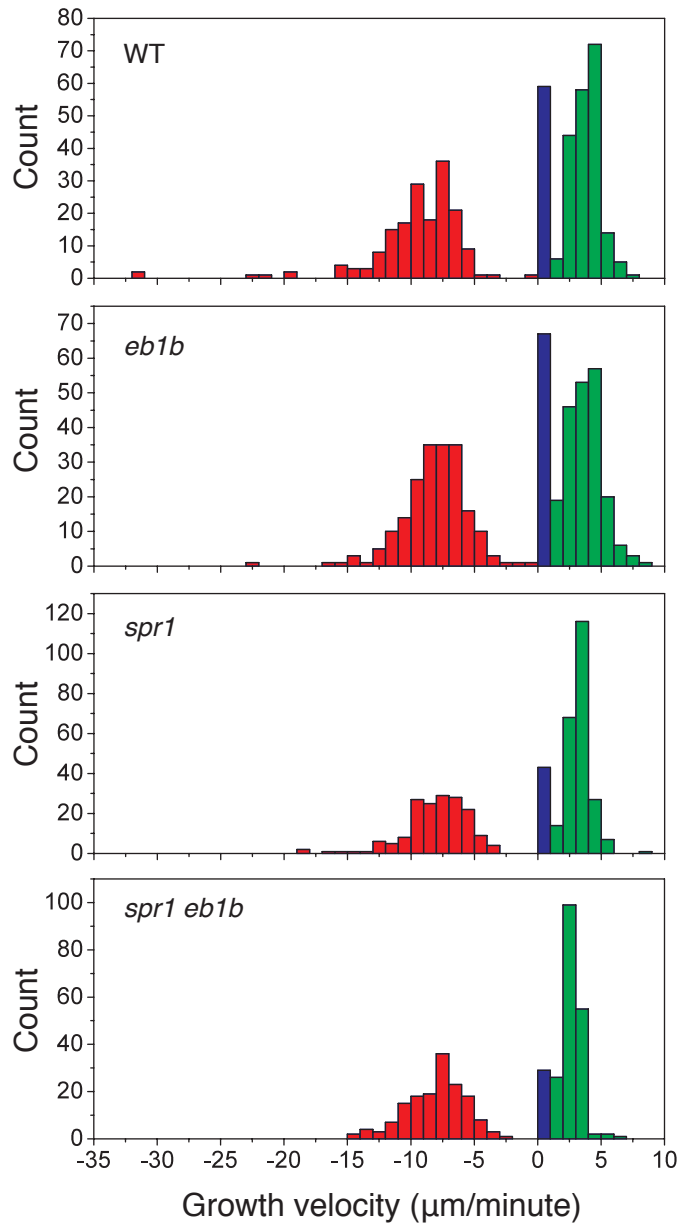
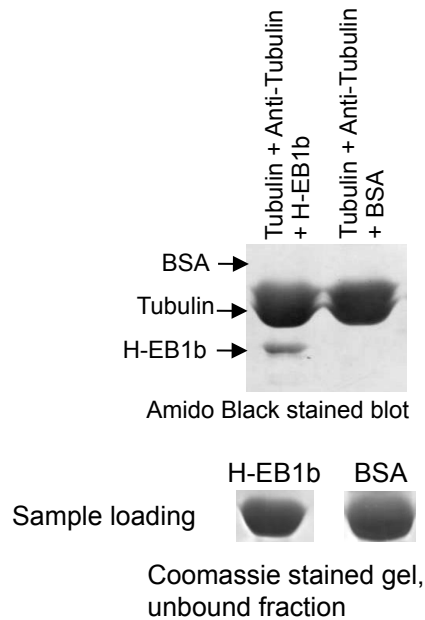


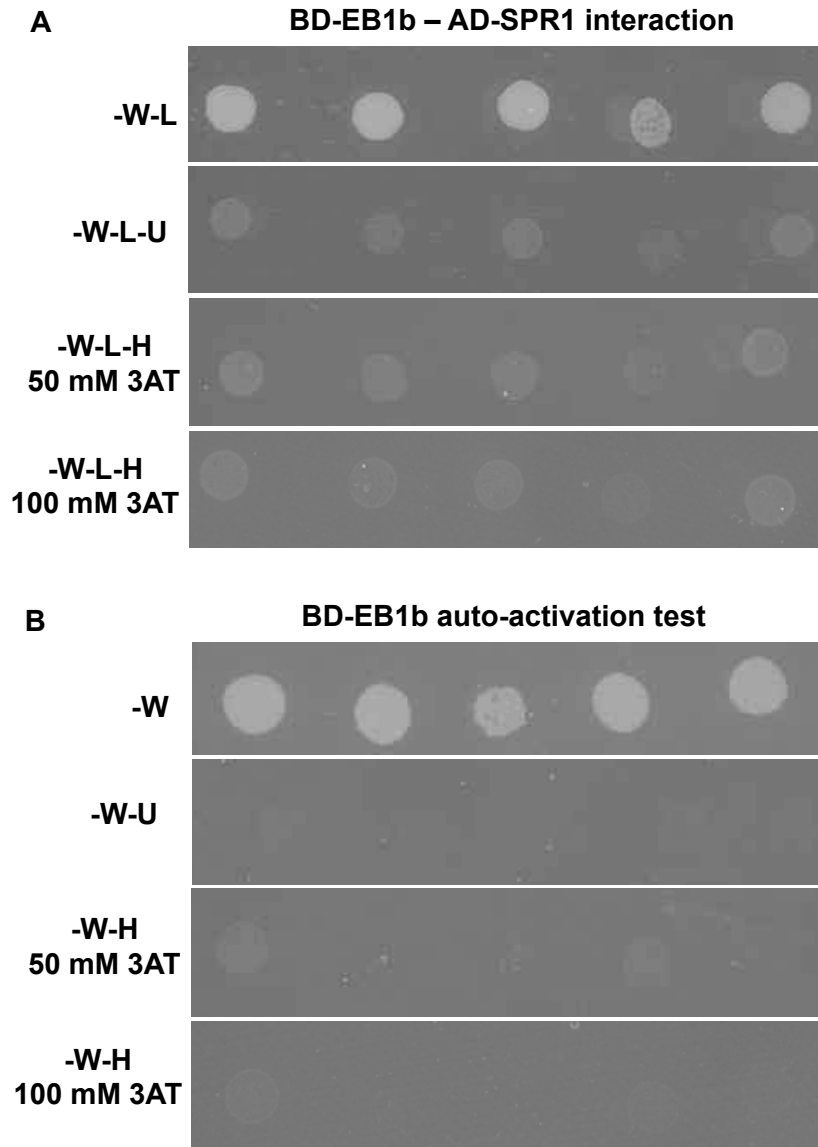
Supplemental Figure 1. Depiction of the *eb1b-2* T-DNA location and determination of related *eb1b-2* gene product formation. **(A)** Diagram of the EB1b gene At5g62500. Solid rectangles and connecting lines represent exons and introns, respectively. Translational start (ATG) and stop sites are noted. The solid triangle represents the T-DNA insertion site in T-DNA line WiscDSLox331A08 (*eb1b-2*), located in intron 6, with the arrow depicting the orientation of primer WiscDSLoxP745 used for T-DNA-genomic flank PCR amplification and DNA sequencing to confirm the location of the insertion site. The arrows below the gene diagram represent the relative locations and orientations of the primers used for RT-PCR analysis. **(B)** Agarose gel images of electrophoresed and ethidium bromide-stained RT-PCR products, revealing the *eb1b-2* transcript likely is truncated compared to wild type. An RT-PCR product comparable to Wild Type (WT) was amplified using *eb1b-2* total RNA as a template and primers EB1BF1 + EB1BR1 (F1R1; 250 bp product) located upstream of the T-DNA insertion site (top image). No RT-PCR product was amplified using *eb1b-2* total RNA and primers EB1BF2 + EB1BR2 (F2R2; 315 bp product) or primers EB1BF1 + EB1BR2 (F1R2; 731 bp product) spanning the insertion site (middle two images). Primers (TUA5F1 + TUA5R1) specific to the TUBULIN ALPHA-5 (TUA5; At5g19780) coding sequence were used in an RT-PCR internal control reaction (bottom image; 560 bp product). **(C)** Film-captured chemiluminescence signal of an immunoblot probed with the anti-EB1 antibody to detect EB1 protein in wild type (lane 1), *eb1b-2* (lane 2), and *eb1a eb1b-2 eb1c* (lane 3; triple mutant control) seedling PAGE-separated extracts. Being that the anti-EB1 antibody cross-reacts with the EB1a protein, it is likely the band visible in the *eb1b-2* sample represents EB1a. EB1a and EB1b are predicted to be 31.1 kD and 32.9 kD in size, respectively. **(D)** Immunoblot probed with the anti-EB1 antibody to detect bacterially expressed and purified H-EB1a (lane 1), H-EB1b (lane 2) and H-EB1c (lane 3) protein electrophoresed in a PAGE gel. Note the antibody more strongly detects H-EB1b compared to H-EB1a, while not detecting H-EB1c.



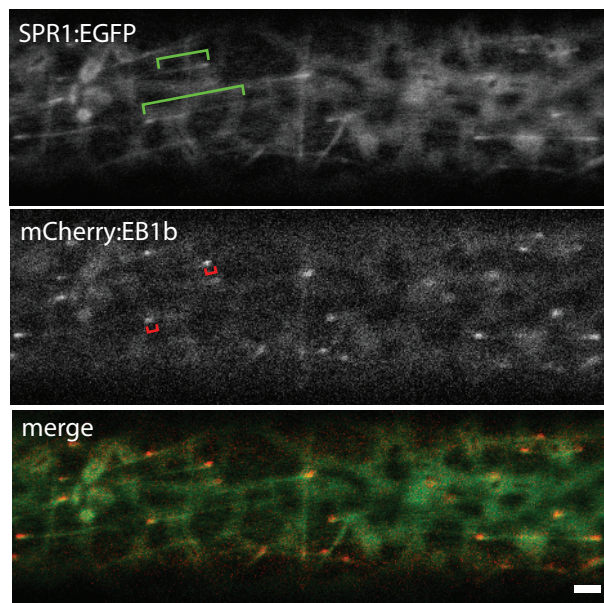
Supplemental Figure 2. Histograms of measured growth velocities of microtubule plus ends. Green (right) - growing ends, red (left) - shrinking ends, blue = paused ends. Mean values for growth and shrinking are compared in figure 4I. The numbers of measured excursions used to acquire each data set were 431, 470, 445 and 37 for wild type, *eb1b-2*, *spr1-6*, and *spr1-6 eb1b-2* respectively. Five cells and plants were sampled per data set.



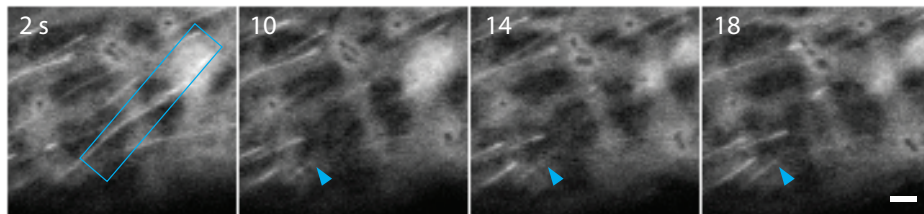
Supplemental Figure 3. *In vitro* pullout of EB1b protein by tubulin heterodimers. Immunoprecipitation assay showing the amount of EB1b protein or Bovine Serum Albumin (BSA; negative control) protein pulled out of solution by Protein A sepharose beads precoated with Anti-tubulin antibody and unpolymerized tubulin. The top image is a Western blot stained with Amido Black to detect the SDS-PAGE-electrophoresed immunoprecipitated protein. Note in lane two that no BSA protein (negative control) co-precipitated with the tubulin. The bottom images are of a coomassie blue-stained SDS-PAGE gel containing electrophoresed protein in the unbound fraction, showing the relative amounts of H-EB1b and BSA protein added to the respective immunoprecipitation solutions. H=6His tag.



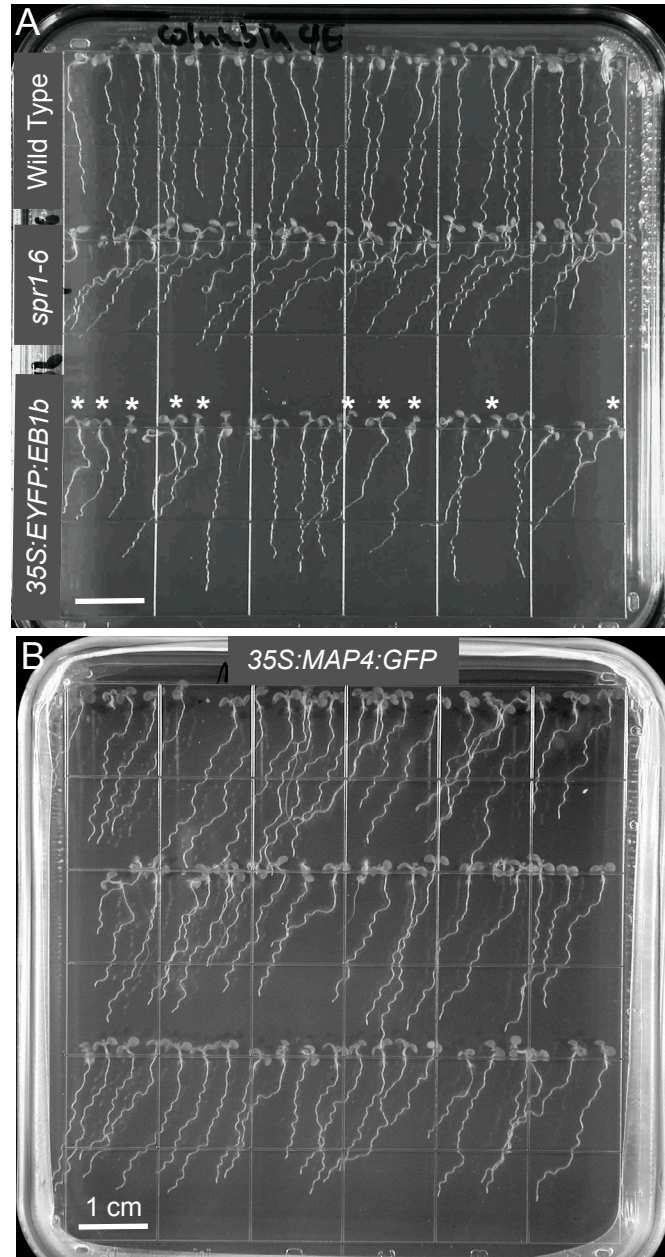
Supplemental Figure 4. Yeast Two-Hybrid analysis of EB1b by SPR1 protein interaction. (A) Shown are five independent co-transformants grown on various yeast dropout media. The EB1b open reading frame was inserted into the *pAS2-1* vector to make a GAL4 DNA binding domain (BD-EB1b) fusion, while the SPR1 open reading frame was inserted into the *pACT2* vector to make a GAL4 DNA activation domain fusion (AD-SPR1). *pAS2-1* harbors the *TRP1* gene, which allows for yeast growth on tryptophan dropout media (-W), while *pACT2* harbors the *LEU2* gene, which allows for yeast growth on leucine dropout media (-L). *MAV203* harbors two separate Gal4 promoter inducible reporter gene constructs (*URA3* and *HIS3*), allowing for redundant testing of bait-prey interactions. Growth on uracil dropout media (-U) as well as on histidine dropout media (-H) signifies that the bait (EB1b) interacts with the prey (SPR1), thereby bringing the BD and AD together to make a functional GAL4 transcription factor. (B) Autoactivation test (negative control). Yeast growth assay showing that the EB1b-GAL4 DNA binding domain fusion (BD-EB1b), by itself in yeast strain *MAV203*, is not able to autoactivate the Gal4p:*URA3* or Gal4p:*HIS3* reporter genes, as shown by little or no yeast growth on respective dropout media. 50 mM or 100 mM 3-amino-1,2,4-triazole (3-AT) was added to the -H media to prevent false positive leaky expression of His3p.



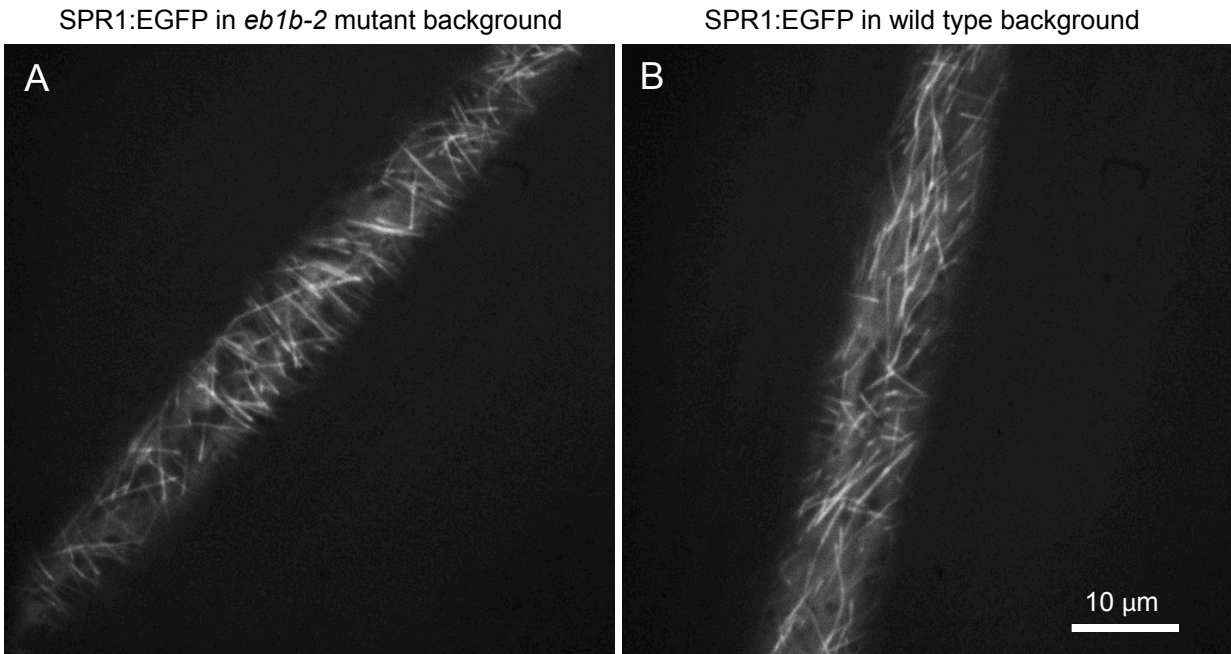
Supplemental Figure 5. Co-localization of pSPR1:SPR1:EGFP and p35S:mCherry:EB1b. Spinning disk confocal images of a dark grown hypocotyl cell (4 day old seedling, upper hypocotyl) co-expressing *SPR1:EGFP* from the *SPR1* promoter and *mCherry:Eb1b* from the *35S* promoter. Images were acquired with approximately 3.5 mWatts of energy as measured at the end of the laser fiber using an 800 msec exposure for each channel and an EM gain of 300. Green (top panel) and red (middle panel) brackets demarcate the extents of SPR1:EGFP and mCherry:EB1b label on microtubules, respectively. Scale bar = 2 microns.



Supplemental Figure 6. Stable boundary of SPR1:EGFP at edge of bleached region. A dark grown hypocotyl cell (4-day-old seedling, upper hypocotyl) was observed at 2 second intervals by laser scanning confocal microscopy using a 256x512 pixel scanned image area. Photobleaching was performed in the area defined by the blue rectangle using two passes of 100% laser power at 488 nm, 200 lines per second. Note that the SPR1:EGFP signal at the lower edge of the bleached region continued to define a fairly sharp boundary with no evidence for movement of signal towards the growing end of the labeled microtubule (upper right of bleached region). Scale bar = 2 microns.



Supplemental Figure 7. Images of seven-day-old Arabidopsis seedlings on 1.5% agar-solidified GM put through the wave assay. Images were taken from below the agar surface (note that root waving images in Figure 1 were taken from above the agar surface). **(A)** Wild Type (top row), *spr1-6* (middle row), and *35S Promoter:EYFP:EB1b* (*35S:EYFP:EB1b*) segregants (bottom row; asterisks denote seedlings expressing EYFP-EB1b). **(B)** *35S Promoter:MAP4 MT BINDING DOMAIN-GFP* (*35S:MAP4:GFP*) expressing seedlings (all three rows). All lines, including wild type, are ecotype Columbia-0. Note that the right root skewing phenotypes of the *35S:MAP4:GFP* and *35S:EYFP:EB1b*-expressing seedlings are comparable to that of *spr1-6* mutant seedlings, raising the possibility that the function of SPR1 or a protein with comparable SPR1 function is compromised in the *35S:MAP4:GFP* and/or *35S:EYFP:EB1b* backgrounds. Scale Bar = 1 cm.



Supplemental Figure 8. SPR1:EGFP localization in an *eb1b-2* hypocotyl cell versus wild type. Confocal images of hypocotyl cells in 3-day-old seedlings expressing SPR1:EGFP in either a homozygous *eb1b-2* mutant background (**A**) or wild type background (**B**). Note there are no obvious differences in SPR1:EGFP tip localization or intensity. Scale bar = 10 microns.