Supplemental material

Lindeboom et al., https://doi.org/10.1083/jcb.201805047

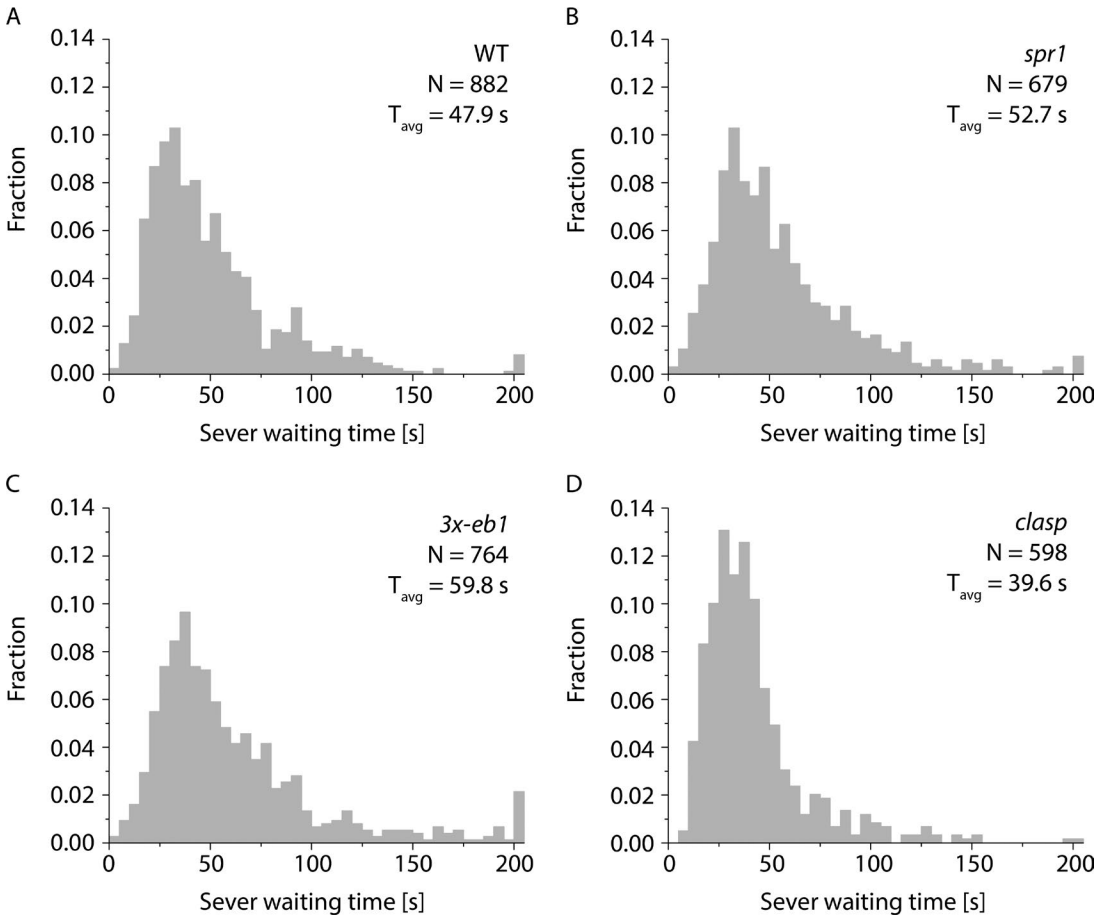


Figure S1. **Sever waiting time distributions and severing preferences. (A–D)** Distribution of sever waiting times from the moment the crossover is formed in WT (A), *spr1* (B), *3x-eb1* (C), and *clasp* (D). Six plants were used for each genotype. Sever waiting times >200 s are added up and shown in the last bin.

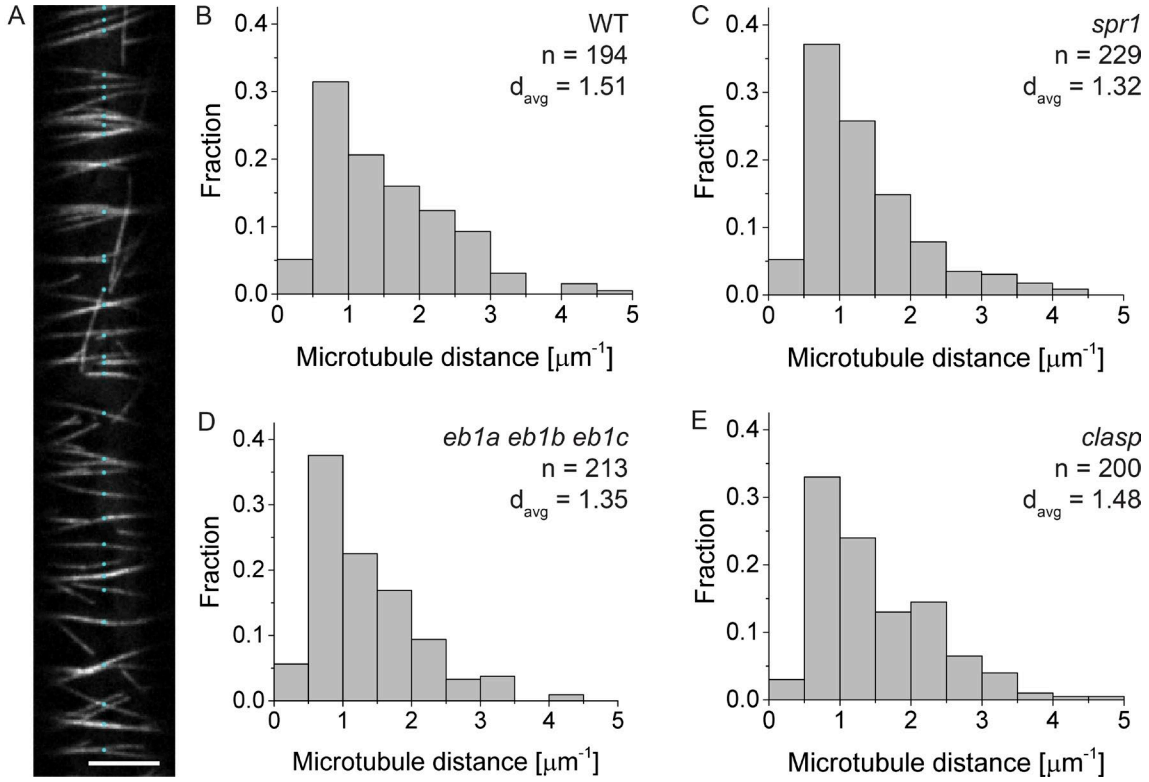


Figure S2. **Longitudinal microtubule distances. (A)** WT example image with detected microtubules labeled in cyan. Scale bar is 5 µm. **(B–E)** Histogram of microtubule distances in WT (B), *spr1* (C), *3x-eb1* (D), and *clasp* (E). Number of detected microtubules indicated by N, d_{avg} is the average distance between microtubules in micrometers, and we used six cells per genotype. A Kruskal–Wallis test followed by a Mann–Whitney *U* test shows that the distribution of bundle distances differs significantly from WT only for *spr1* ($P < 0.05$).

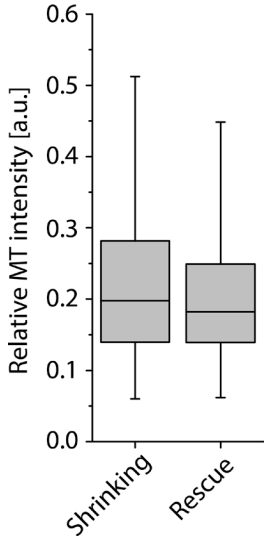


Figure S3. **Microtubule signal does not positively correlate with rescue events.** Boxplot of relative MT signal intensity on microtubules that continue shrinking and microtubules that get rescued. Boxplots show the 25th and 75th percentile as box edges, the line in the box indicates median value, and the whiskers show the 2.5th and 97.5th percentile. Microtubules were observed shrinking in 2,716 frames, and we observed rescue 301 times. The relative MT signal was not significantly higher for instances of microtubule rescue ($P > 0.98$, Mann–Whitney *U* test).

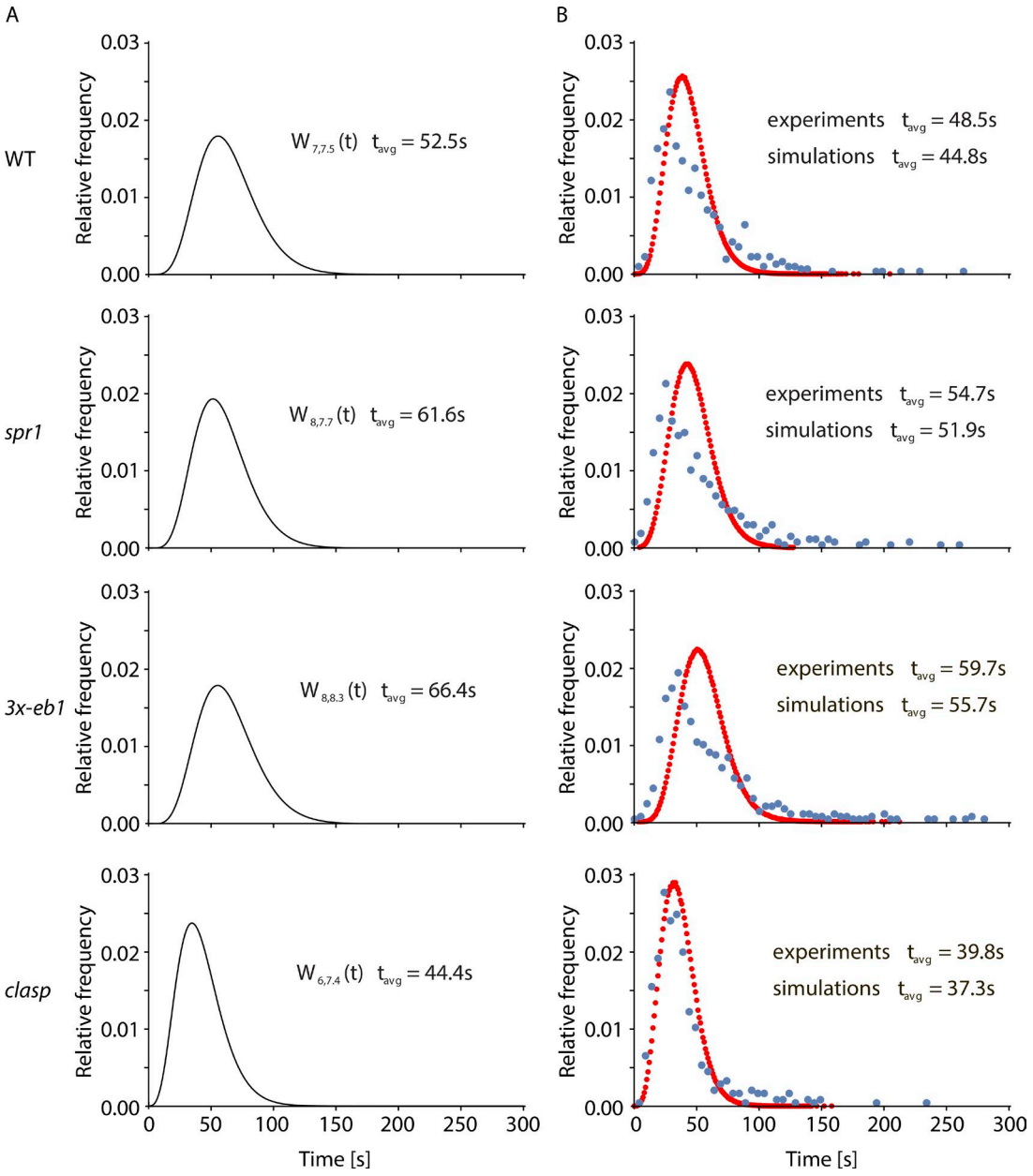


Figure S4. **Observed and fitted sever waiting time distributions. (A and B)** The optimal intrinsic severing waiting time distribution used in simulations (A) and the comparison between computed conditional severing waiting time distribution (red dots) and experimentally measured distribution (blue dots), for WT, *spr1*, *3x-eb1*, and *clasp* (B).

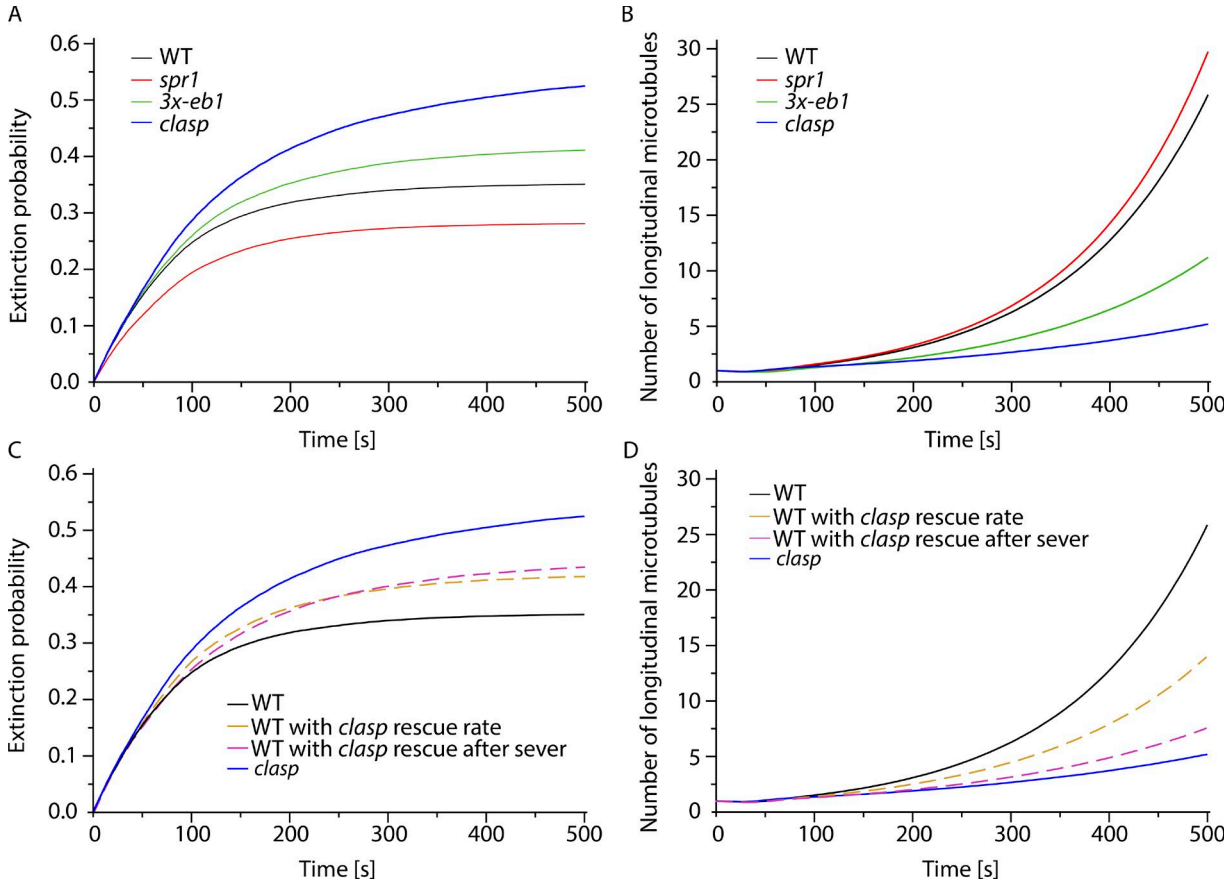
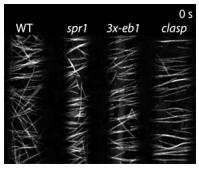
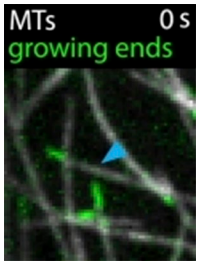


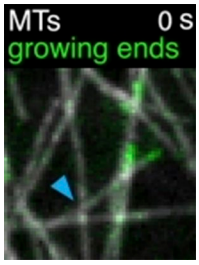
Figure S5. **Number of longitudinal microtubules in the simulations after 500 s as *Ps,+* and *rr* are varied.** Blue triangles represent WT microtubules with changed rescue after severing probability, respectively: $P_{s,+}$ = 0, $P_{s,+}$ = 0.025, $P_{s,+}$ = 0.05, $P_{s,+}$ = 0.075, $P_{s,+}$ = 0.1, $P_{s,+}$ = 0.125, $P_{s,+}$ = 0.15, $P_{s,+}$ = 0.175, $P_{s,+}$ = 0.2, $P_{s,+}$ = 0.225, and $P_{s,+}$ = 0.25. Red dots represent WT microtubules with changed intrinsic rescue rate, respectively: r_r = 0.76, r_r = 0.89, r_r = 1.03, r_r = 1.16, r_r = 1.30, r_r = 1.43, r_r = 1.56, r_r = 1.69, r_r = 1.83, r_r = 1.96, and r_r = 2.09 events/min.



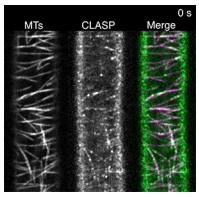
Video 1. **Blue light–induced microtubule reorientation in dark-grown hypocotyl epidermal cells expressing YFP-TUA5 in WT, *spr1*, *3x-eb1*, and *clasp* genetic backgrounds.** Playback rate is 30 frames/second (FPS).



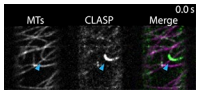
Video 2. **Example of crossover severing event in a WT dark-grown hypocotyl epidermal cell that generates a shrinking new plus end.** The cyan arrowhead marks the microtubule crossover site. The yellow > marks the new plus end. Growing microtubule ends are shown in green. Playback rate is 7 FPS.



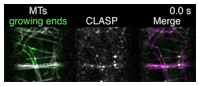
Video 3. **Example of crossover severing event in a WT dark-grown hypocotyl epidermal cell that generates a growing new plus end.** The cyan arrowhead marks the microtubule crossover site. The yellow > marks the new plus end. Growing microtubule ends are shown in green. Playback rate is 7 FPS.



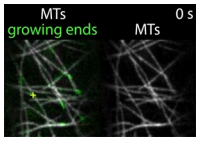
Video 4. **Time-lapse video of CLASP and MT colocalization in dark grown hypocotyl epidermal cell in a *clasp* mutant rescued with a YFP-CLASP construct and coexpressing mCherry-TUA5.** Playback rate is 15 FPS.



Video 5. **Example time-lapse images showing strong CLASP label on highly curved microtubules.** Highly curved microtubules are marked by a cyan arrowhead. The example video features a dark-grown epidermal hypocotyl cell of a *clasp* mutant rescued with a YFP-CLASP construct and coexpressing mCherry-TUA5. Playback rate is 15 FPS.



Video 6. **Example of crossover severing event in a WT dark-grown hypocotyl epidermal cell that generates a growing new plus end.** The cyan arrowhead marks the microtubule crossover site. The yellow > marks the new plus end. Growing microtubule ends are shown in green on the left panel, and the rest of the microtubule lattice is shown in grayscale. The middle panel shows the CLASP signal, and the right panel shows a merge of the microtubule and CLASP signal. Playback rate is 7 FPS.



Video 7. **Example of a section of a WT cell expressing YFP-TUA5 dark-grown hypocotyl cell where all the identified crossovers are marked with yellow plus signs.** Growing microtubule ends are shown in green. Playback rate is 30 FPS.

Provided online are two tables in Excel. Table S1 lists the parameter values used for modeling of microtubule amplification, and Table S2 lists the sequences of primers used in cloning.