

Figure S1. **Silencing Sec10 results in a growth defect.** Related to Fig. 3. **(A)** Chlorophyll autofluorescence images of 7-d-old plants regenerated from protoplasts. Bar, 150 μ m. **(B)** Plant area was quantified by measuring the area of chlorophyll autofluorescence. All data are normalized to the wild-type control. Error bars represent SEM. Numbers in the bars indicate number of plants analyzed for each condition. Letters above the bars indicate statistical groups with $\alpha < 0.05$ from ANOVA.

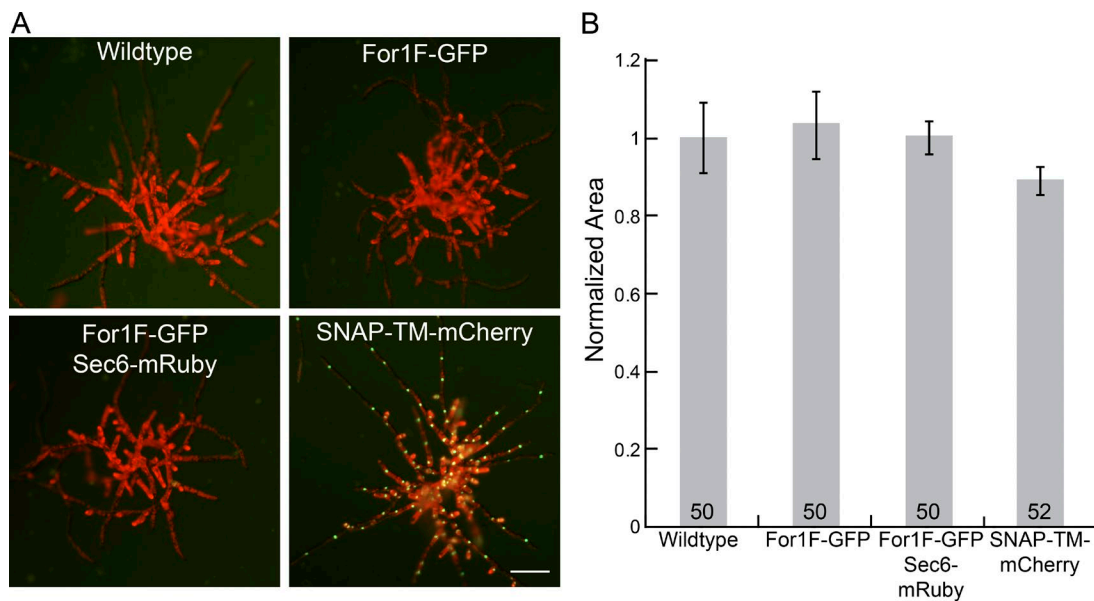


Figure S2. **Stable lines grow similar to wild type.** Related to Figs. 3, 4, and 5. **(A)** Chlorophyll autofluorescence images of 7-d-old plants regenerated from protoplasts. Bar, 150 μ m. **(B)** Plant area was quantified by measuring the area of chlorophyll autofluorescence. All data are normalized to the wild-type control. Error bars represent SEM. Numbers in the bars indicate number of plants analyzed for each condition. ANOVA grouped all samples into the same statistical group.

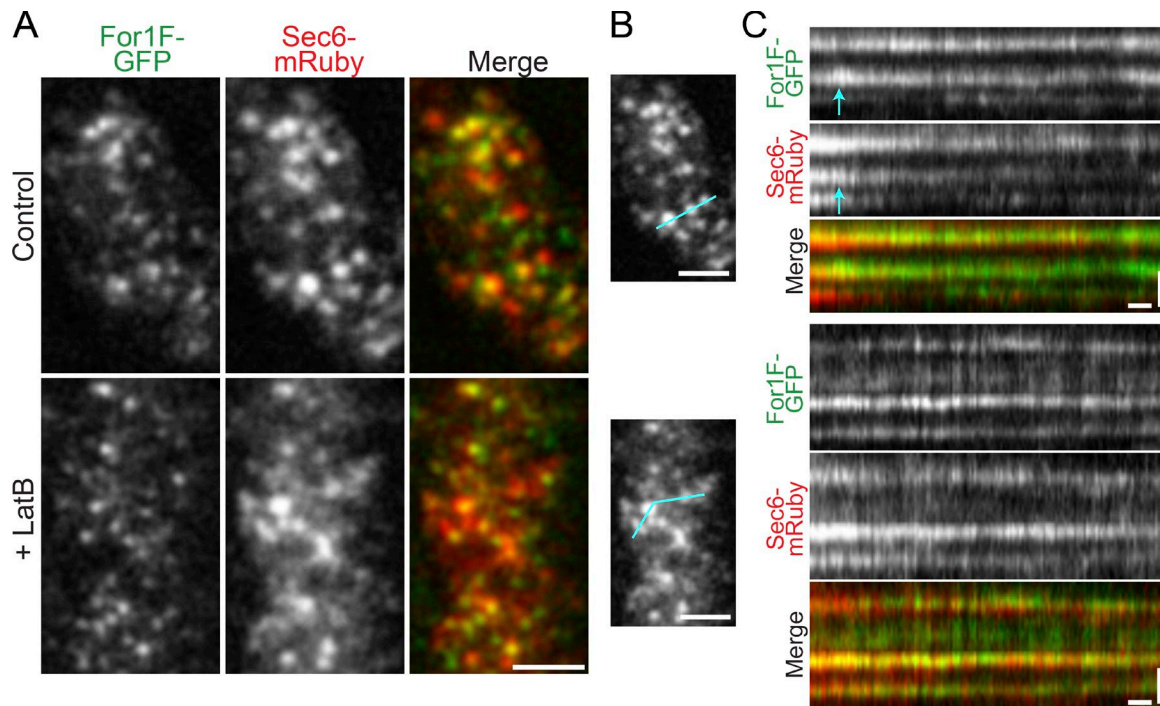


Figure S3. **Rapid dissociation of Sec6-mRuby depends on actin.** Related to Fig. 4 (C and E). **(A)** VAEM images of the cortex of cells expressing endogenously tagged For1F-GFP and Sec6-mRuby in the presence (control) or absence (LatB) of actin filaments. Bar, 2 μ m. Also see Videos 2 and 5. **(C)** Kymograph analysis of For1F-GFP and Sec6-mRuby dynamics from the control and LatB-treated cells in B. Bars: (horizontal) 2 s; (vertical) 1 μ m. The line used for the kymograph is shown in the image to the left of the kymographs. Bar, 2 μ m.

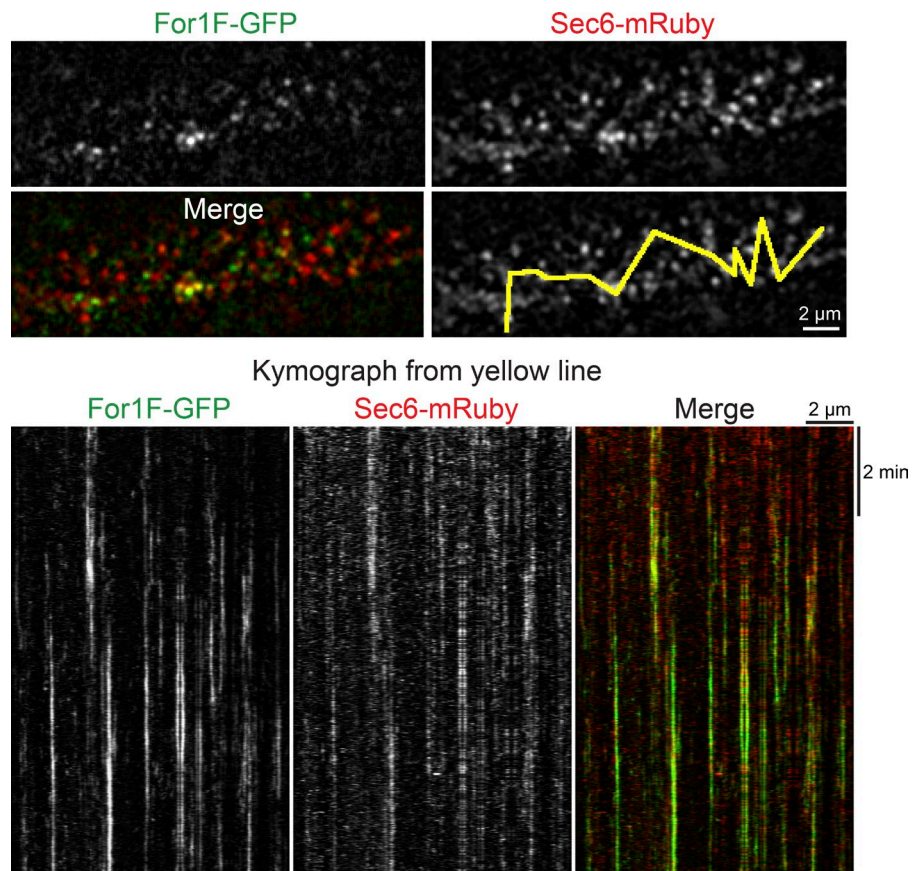
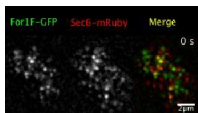
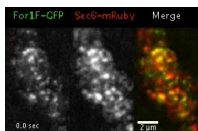


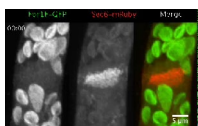
Figure S4. **Quantification of cortical particle interaction through time.** Related to Fig. 4 F. A line was drawn through the brightest Sec6-mCherry particles in the first frame of a 10-min time-lapse acquisition. Kymographs from this line (3 pixels wide) in both the Sec6-mCherry and For1F-GFP channels were generated in Fiji. The PCC was calculated with the Coloc2 plug-in for Fiji. The PCCs for all kymographs are reported in Fig. 4 F.



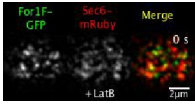
Video 1. **Simultaneous VAEM imaging of cortical For1F-GFP and Sec6-mRuby.** Total acquisition time: 10 min. Related to Fig. 4 C.



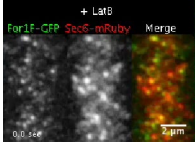
Video 2. **Simultaneous VAEM imaging of cortical For1F-GFP and Sec6-mRuby.** Total acquisition time: 30 s. Related to Fig. S3.



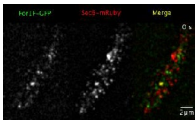
Video 3. **Laser scanning confocal imaging of For1F-GFP and Sec6-mRuby during cell division.** Images are maximum projections of a confocal z-stack. Related to Fig. 5 B.



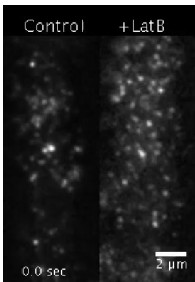
Video 4. **Simultaneous VAEM imaging of cortical For1F-GFP and Sec6-mRuby treated with LatB.** Total acquisition time: 10 min. Related to Fig. 4 C.



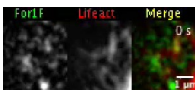
Video 5. **Simultaneous VAEM imaging of cortical For1F-GFP and Sec6-mRuby treated with LatB.** Total acquisition time: 30 s. Related to Fig. S3.



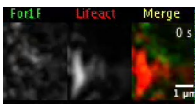
Video 6. **Simultaneous VAEM imaging of cortical For1F-GFP and CLC-mRuby.** Related to Fig. 4 D.



Video 7. **VAEM imaging of cortical For1F-GFP.** Related to Fig. 6 A.



Video 8. **Simultaneous VAEM imaging of cortical For1F-GFP and Lifeact-mCherry.** Related to Fig. 6 E.



Video 9. **Simultaneous VAEM imaging of cortical For1F-GFP and Lifeact-mCherry.** Related to Fig. 6 G.

Provided online is Table S1 in Excel showing the primers used in this study.