

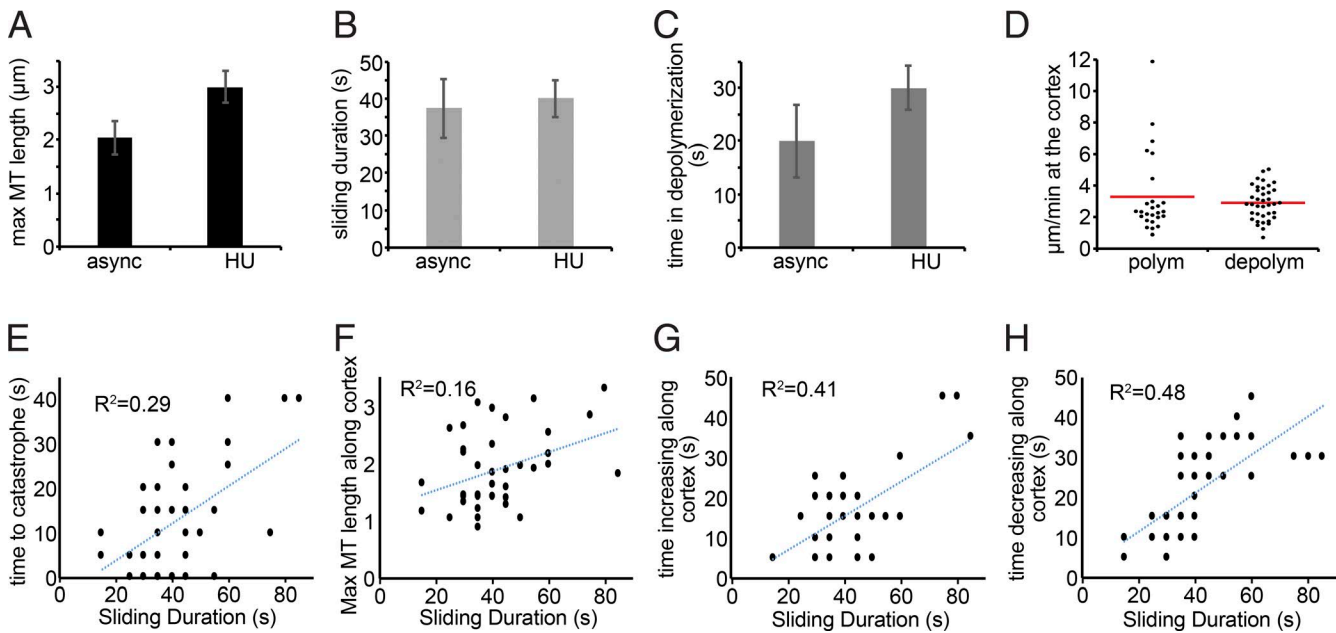
Estrem et al., <https://doi.org/10.1083/jcb.201611105>

Figure S1. **Properties of the microtubule while sliding along the cell cortex.** (A) Median of the maximum astral microtubule (MT) length reached during a sliding event in asynchronous (async) pre-anaphase cells and HU-arrested cells. (B) Median sliding duration in seconds. (C) Median time spent in depolymerization during a sliding event. (A–C) Pre-anaphase cells, $n = 28$; HU-arrested cells, $n = 39$. Error bars are 95% c.i. (D) Dot plot of the rate of polymerization (polym; $n = 26$ microtubules) and depolymerization (depolym; $n = 39$ microtubules) of microtubules along the cortex during sliding events. (E) Scatter plot depicting the coefficient of determination for time to catastrophe (seconds) and sliding duration (seconds). $n = 32$ microtubules. (F) Scatter plot depicting the coefficient of determination for maximum length of microtubules along the cortex (μm) and sliding duration (seconds). $n = 34$ microtubules. (G) Scatter plot depicting the coefficient of determination for time the microtubule spends increasing its length along the cortex (seconds) and sliding duration (seconds). $n = 25$ microtubules. (H) Scatter plot depicting the coefficient of determination for time the microtubule spends decreasing its length along the cortex (seconds) and sliding duration (seconds). $n = 30$ microtubules.

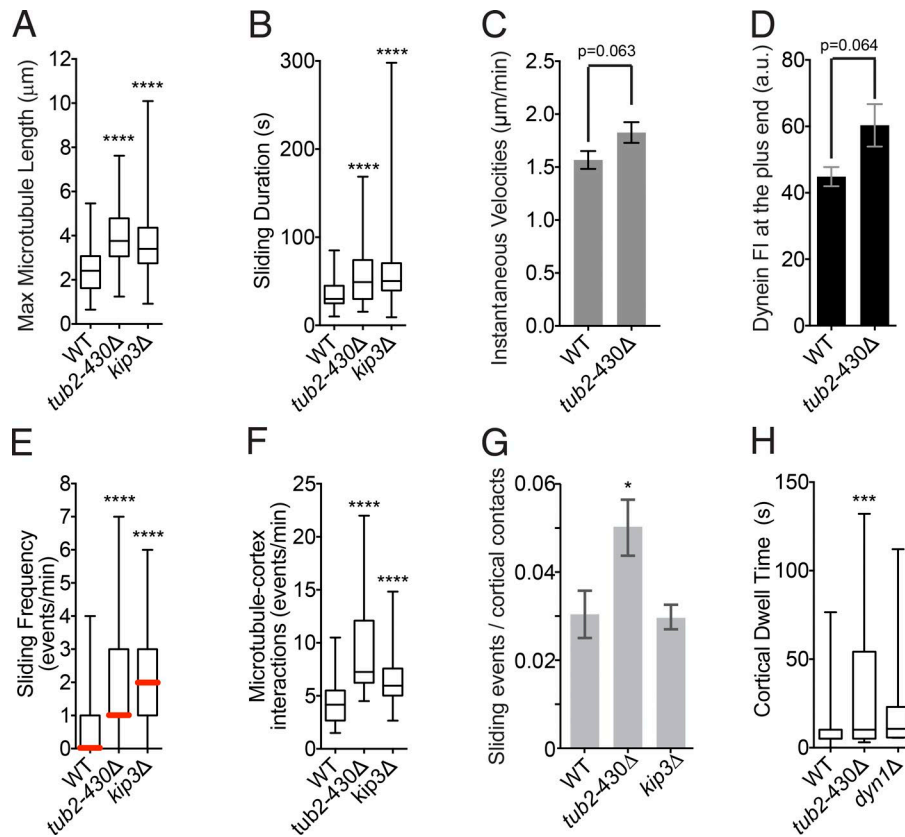


Figure S2. **Mutants with long microtubules increase cortical contacts, sliding frequency, and sliding duration.** (A) Box and whisker plot of maximum microtubule length reached during a sliding event (μm). The center bar denotes the median, the box denotes the first and third quartiles, and whiskers are maxima and minima. WT, $n = 51$ sliding events; *tub2-430 Δ* , $n = 97$; *kip3 Δ* , $n = 208$. (B) Box and whisker plot of sliding event duration (seconds). WT, $n = 52$ sliding events; *tub2-430 Δ* , $n = 97$; *kip3 Δ* , $n = 204$. (C) Mean instantaneous velocities ($\mu\text{m}/\text{min}$) for spindle movement in pre-anaphase cells. WT, $n = 95$ cells; *tub2-430 Δ* , $n = 144$. Error bars are SEM. (D) Mean fluorescent intensity (arbitrary units [a.u.]) of Dyn1-3GFP foci (FI) at the plus end of the microtubule. WT, $n = 68$ microtubule plus ends; *tub2-430 Δ* , $n = 106$. Error bars are SEM. (E) Box and whisker plot of sliding frequency (sliding events/min). Red bars denote the median, boxes denote the first and third quartiles, and whiskers are maxima and minima. WT, $n = 91$ cells; *tub2-430 Δ* , $n = 73$; *kip3 Δ* , $n = 53$. (F) Box and whisker plot of microtubule–cortex interactions (events/min). WT, $n = 39$ cells; *tub2-430 Δ* , $n = 38$; *kip3 Δ* , $n = 57$. (G) Mean value for the ratio of sliding events over cortical contacts. WT, $n = 39$ cells; *tub2-430 Δ* , $n = 38$; *kip3 Δ* , $n = 57$. Error bars are SEM. (H) Box and whisker plot of cortical dwell time (seconds). WT, $n = 52$ cells; *tub2-430 Δ* , $n = 49$; *dyn1 Δ* , $n = 64$. Asterisks denote a significant difference from WT. *, $P < 0.05$; **, $P < 0.005$; ***, $P < 0.0006$; ****, $P < 0.0001$; determined by *t* test with the exception of G where the $P < 0.05$ was determined by Fisher's exact test.

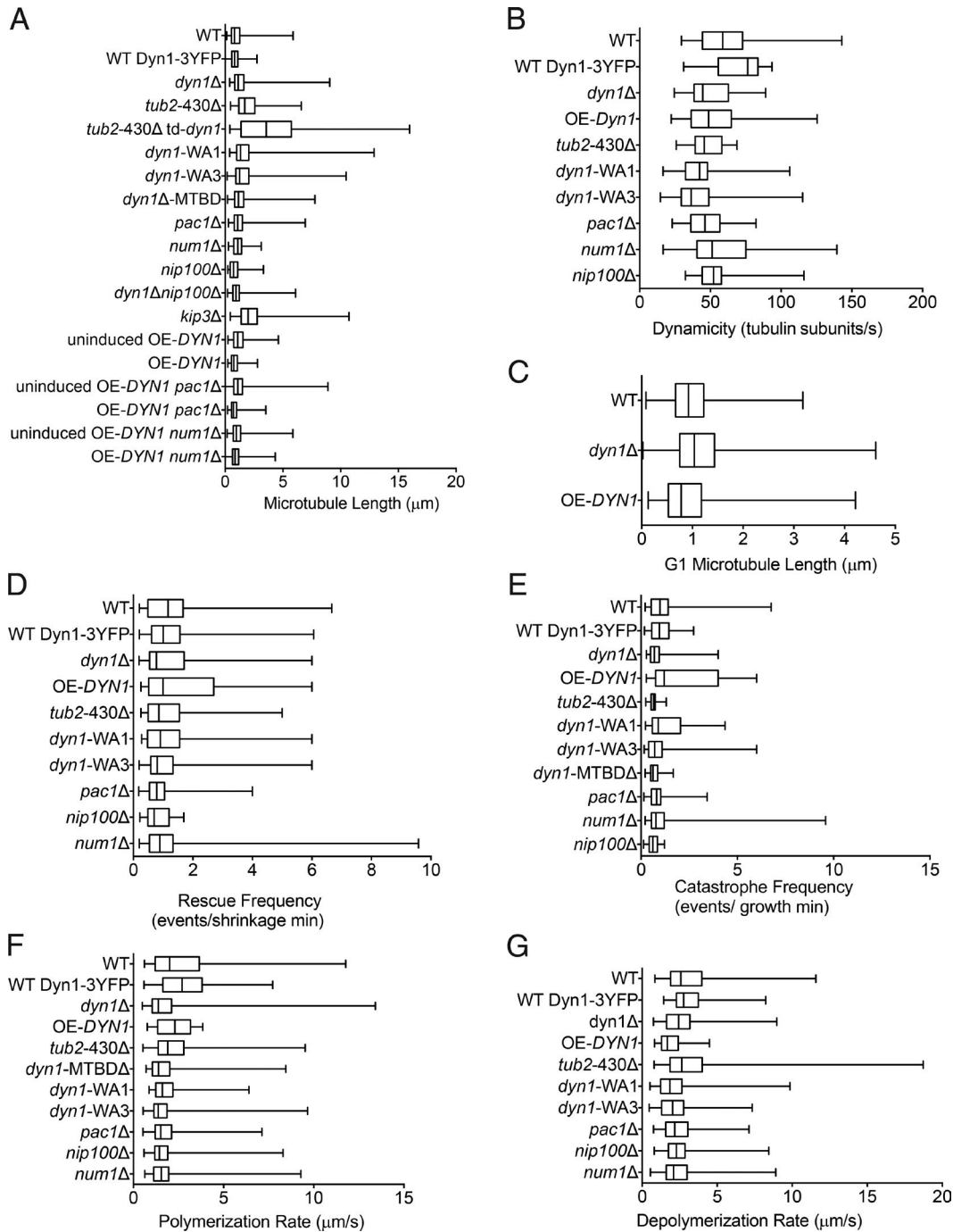


Figure S3. **Distributions of microtubule dynamics measurements.** (A) Box and whisker plot of astral microtubule length (μm) of cells in pre-anaphase. The center bar denotes the median, the box denotes the first and third quartiles, and whiskers are maxima and minima. WT, $n = 371$ microtubules; *Dyn1-3YFP*, $n = 300$; *dyn1Δ*, $n = 278$; *tub2-430Δ*, $n = 144$; *tub2-430Δ td-dyn1*, $n = 46$; *dyn1-WA1*, $n = 175$; *dyn1-WA3*, $n = 184$; *dyn1Δ-MTBD*, $n = 158$; *pac1Δ*, $n = 160$; *num1Δ*, $n = 203$; *nip100Δ*, $n = 134$; *dyn1Δnip100Δ*, $n = 1,310$; *kip3Δ*, $n = 132$; uninduced OE-DYN1, $n = 208$; OE-DYN1, $n = 339$; uninduced OE-DYN1 *pac1Δ*, $n = 1,048$; OE-DYN1 *pac1Δ*, $n = 753$; uninduced OE-DYN1 *num1Δ*, $n = 1,662$; OE-DYN1 *num1Δ*, $n = 907$. (B) Box and whisker plot of dynamicity (tubulin subunits/second). WT, $n = 68$ growth or shrinkage events; DYN1-3YFP, $n = 22$; *dyn1Δ*, $n = 47$; OE-DYN1, $n = 48$; *tub2-430Δ*, $n = 16$; *dyn1-WA1*, $n = 52$; *dyn1-WA3*, $n = 87$; *pac1Δ*, $n = 60$; *num1Δ*, $n = 76$; *nip100Δ*, $n = 22$. (C) Box and whisker plot of astral microtubule length in G1 cells. WT, $n = 528$; *dyn1Δ*, $n = 843$; OE-DYN1, $n = 878$. (D) Box and whisker plot of rescue frequency (events/shrinkage minute). WT, $n = 68$ microtubules; DYN1-3YFP, $n = 22$; *dyn1Δ*, $n = 47$; OE-DYN1, $n = 48$; *tub2-430Δ*, $n = 55$; *dyn1-WA1*, $n = 52$; *dyn1-WA3*, $n = 87$; *pac1Δ*, $n = 60$; *num1Δ*, $n = 76$; *nip100Δ*, $n = 23$. (E) Box and whisker plot of catastrophe frequency (events/growth minute). WT, $n = 68$ microtubules; DYN1-3YFP, $n = 22$; *dyn1Δ*, $n = 47$; OE-DYN1, $n = 48$; *tub2-430Δ*, $n = 55$; *dyn1-WA1*, $n = 52$; *dyn1-WA3*, $n = 87$; *dyn1-MTBD-3YFP*, $n = 60$; *num1Δ*, $n = 76$; *nip100Δ*, $n = 23$. (F) Box and whisker plot of polymerization rate ($\mu\text{m}/\text{second}$). WT, $n = 68$ microtubules; DYN1-3YFP, $n = 22$; *dyn1Δ*, $n = 47$; OE-DYN1, $n = 48$; *tub2-430Δ*, $n = 55$; *dyn1-WA1*, $n = 52$; *dyn1-WA3*, $n = 87$; *dyn1Δ-MTBD*, $n = 31$; *pac1Δ*, $n = 60$; *num1Δ*, $n = 76$; *nip100Δ*, $n = 23$. (G) Box and whisker plot of depolymerization rate ($\mu\text{m}/\text{second}$). WT, $n = 68$ microtubules; DYN1-3YFP, $n = 22$; *dyn1Δ*, $n = 47$; OE-DYN1, $n = 48$; *tub2-430Δ*, $n = 55$; *dyn1-WA1*, $n = 52$; *dyn1-WA3*, $n = 87$; *pac1Δ*, $n = 60$; *num1Δ*, $n = 76$; *nip100Δ*, $n = 23$.

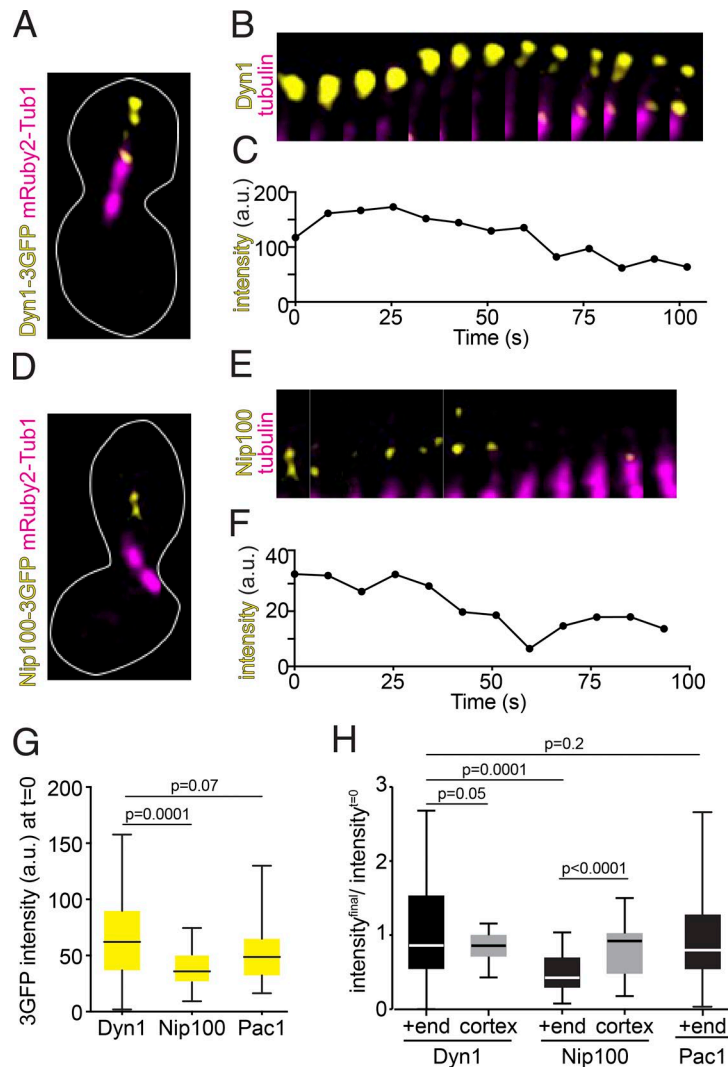
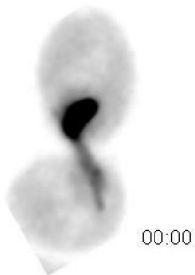
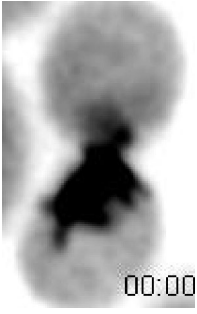


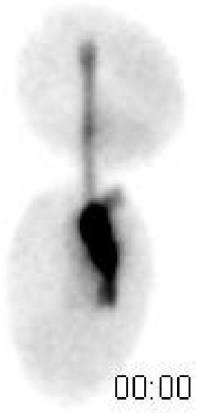
Figure S4. Dynein remains at the plus end of the microtubule during sliding, whereas the dynactin component Nip100 is depleted. (A) Representative image of a cell expressing Dyn1-3GFP (yellow) and mRuby2-Tub1 (magenta). The image is a maximum intensity projection from a confocal Z series. (B) Time series of images showing Dyn1-3GFP localization at the plus end of the microtubule during a sliding event in B. a.u., arbitrary units. $n = 13$ measurements during 110 s. (D) Representative image of a cell expressing Nip100-3GFP (yellow) and mRuby2-Tub1 (magenta). The image is a maximum intensity projection from a confocal Z series. (E) Time series of images showing Nip100-3GFP localization at the plus end of the microtubule during a sliding event. (F) Fluorescence intensity measurements of the Nip100-3GFP at the plus end of the microtubule during the sliding event in E. $n = 12$ measurements during 95 s. (G) Box and whisker plot of 3GFP fluorescent intensity (arbitrary units) at the first time point of a sliding event. The center bar denotes the median, the box denotes the first and third quartiles, and whiskers are maxima and minima. Dyn1, $n = 43$ sliding events; Nip100, $n = 28$; Pac1, $n = 28$. (H) Box and whisker plot of the ratios of fluorescence intensity at the final time point of the sliding event divided by the intensity at the first time point. Black boxes represent plus-end measurements. Gray boxes represent measurements of cortical foci. Dyn1, $n = 37$ time-matched ratio values for plus end and cortical foci; Nip100, $n = 30$; Pac1, $n = 28$. P-values were determined by t test.



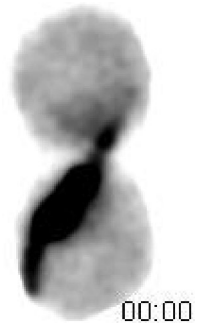
Video 1. Tub1-GFP dynamics in a WT cell. Time-lapse images of a WT cell expressing Tub1-GFP were captured on a spinning-disk confocal microscope at 5-s intervals for 10 min. Each image represents a composite of 11 planes separated by 600 nm. The video plays at seven frames per second. Strain: WT, yJM0562.



Video 2. **Tub1-GFP dynamics in a *dyn1Δ* cell.** Time-lapse images of a *dyn1Δ* cell expressing Tub1-GFP were captured on a spinning-disk confocal microscope at 9-s intervals for 10 min. Each image represents a composite of 19 planes separated by 300 nm. The video plays at seven frames per second. Strain: *dyn1Δ*, yJM1022.



Video 3. **Tub1-GFP dynamics in a *tub2-430Δ* cell.** Time-lapse images of a *tub2-430Δ* cell expressing Tub1-GFP were captured on a spinning-disk confocal microscope at 9-s intervals for 10 min. Each image represents a composite of 19 planes separated by 300 nm. The video plays at seven frames per second. Strain: *tub2-430Δ*, yJM0301.



Video 4. **Tub1-GFP dynamics in a *kip3Δ* cell.** Time-lapse images of a *kip3Δ* cell expressing Tub1-GFP were captured on a spinning-disk confocal microscope at 9-s intervals for 10 min. Each image represents a composite of 19 planes separated by 300 nm. The video plays at seven frames per second. Strain: *kip3Δ*, yJM1345.

Table S1 is available as an Excel file and shows the strains used in this study.