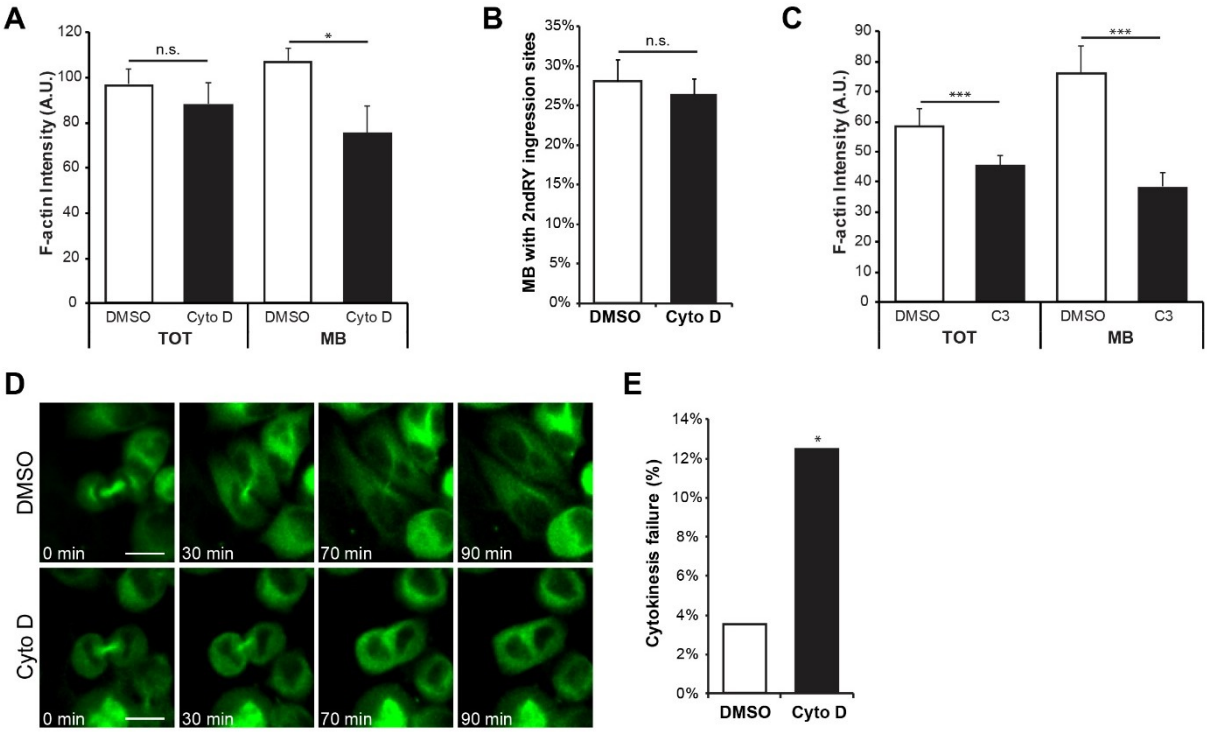
Supplemental Figures

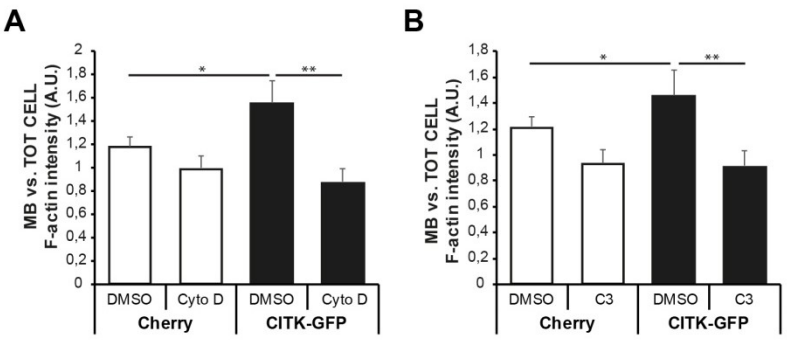


Supplemental Figure 1. Effects of F-actin depolymerisation in late cytokinesis. (A) HeLa cells were synchronized in late cytokinesis, treated with 0,4 μM Cytochalasin D (CytoD) or DMSO for 60 min and immunostained for α-tubulin and F-actin. Total cell (*TOT*) or midbody (*MB*) F-actin intensity was quantified. (B) Percentage of midbodies with secondary (*2ndRY*) ingression sites in cell treated as in (A) (n > 30) (C) HeLa cells were synchronized in late cytokinesis, treated with the RhoA inhibitor C3-transferase or control solution for 60 min and immunostained for α-tubulin and F-actin. Total cell (*TOT*) or midbody (*MB*) F-actin intensity was quantified (n > 30, 3 independent experiments).

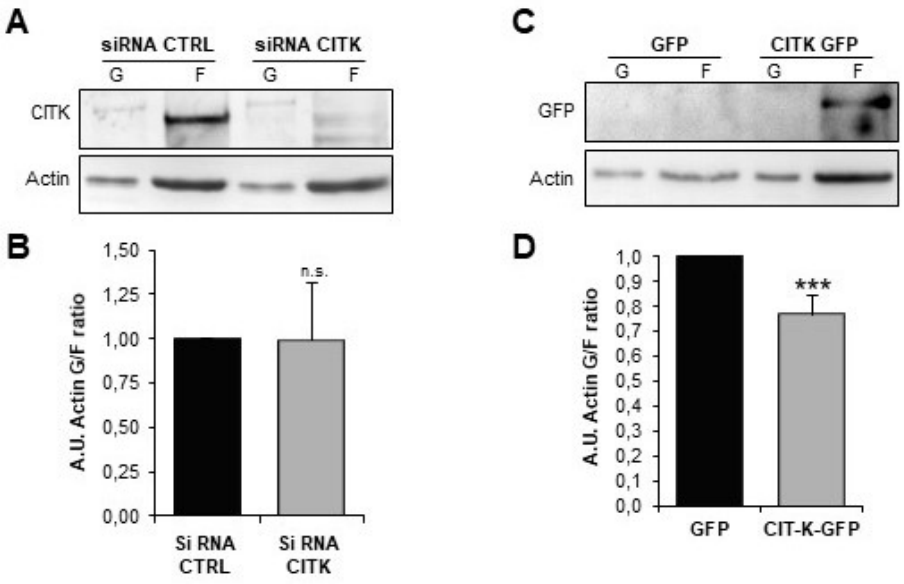
Data shown in histograms are means ± s.e.m. Statistical significance was assessed using a two tails Student T-test. * $p < 0.05$. *** $p < 0.001$.

(D) Time-lapse imaging of α-tubulin-GFP-expressing HeLa cells was started after Cytochalasin D addition. Scale bars, 20μm. For full movie, see Movies 1-2. (C) Quantification of cytokinesis failures of cells treated as in (D), only cells with complete cleavage furrow ingression at the time of Cytochalasin D addition were analyzed.

Data shown in histogram are the percentage of cytokinesis failures on the total number of cytokinesis analyzed. Statistical significance was assessed using a Chi-square test. * $p < 0.05$ (n > 60).

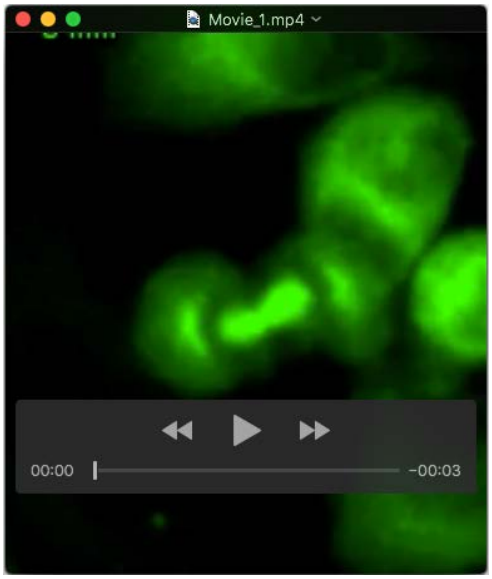


Supplemental Figure 2. Cytochalasin D treatment and RhoA inhibition could revert actin stabilization at the midbody caused by CITK overexpression. HeLa cells expressing either GFP or CITK-GFP were synchronized in late cytokinesis, treated with 0.4 μM Cytochalasin D or DMSO (A) and with RhoA inhibitor C3-transferase or control solution (B) for 60 min and immunostained for α-tubulin and F-actin. Midbody (*MB*) versus total cell (*TOT*) F-actin intensity was quantified (n > 30, 6 independent experiments).
Data shown in histograms are means ± s.e.m. Statistical significance was assessed using a two tails Student T-test. * $p < 0.05$. ** $p < 0.01$.

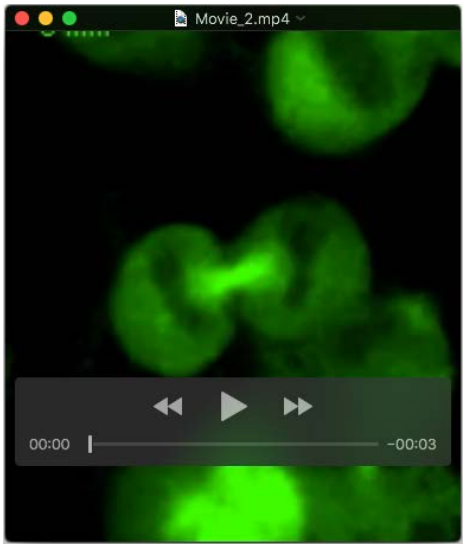


Supplemental Figure 3. CITK overexpression increases actin polymerization. G/F actin fractionation was performed on HeLa cells treated with control or CITK siRNAs (A, B) and on GFP or CITK-GFP expressing HeLa cells (C, D) and the resulting lysates were run on western blot, then G/F actin ratio was calculated (n =4). Data shown in histograms are means ± s.e.m. Statistical significance was assessed using a two tailed Student T-test. *** $P < 0.001$ n.s. not significant.

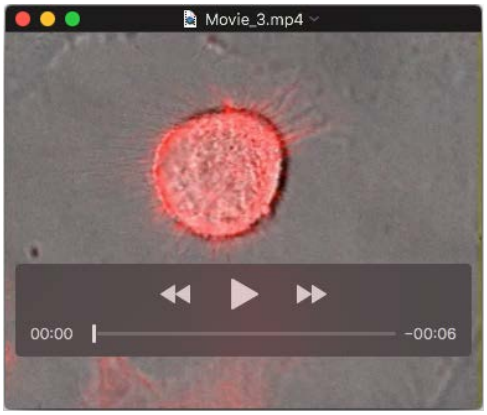
Movies



Movie 1. Time-lapse of HeLa cells stably expressing TUB-GFP synchronized and treated with DMSO after midbody formation.

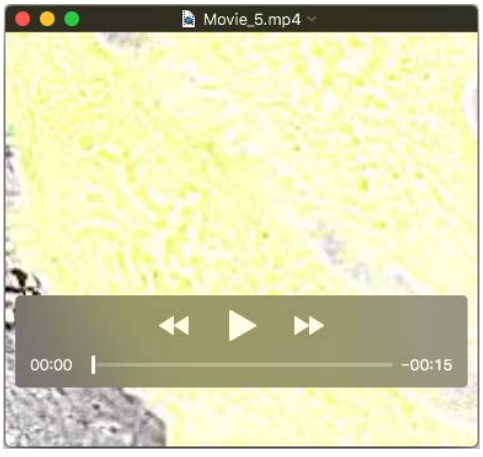


Movie 2. Time-lapse of HeLa cells stably expressing TUB-GFP synchronized and treated with 1uM Cytochalasin D after midbody formation.

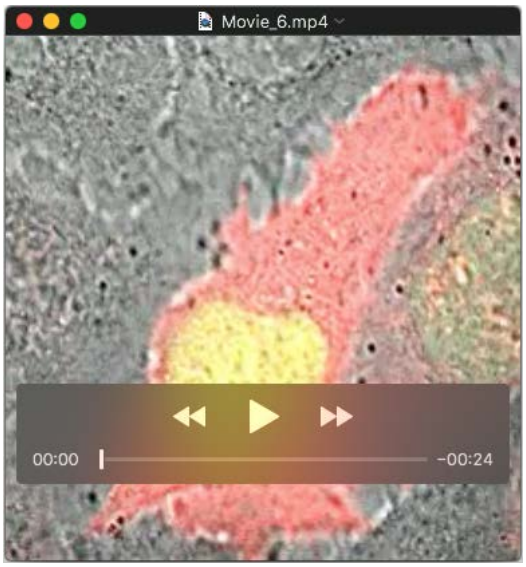


Movie 3. Time-lapse of HeLa cells transfected with Lifeact-RFP treated with control siRNA.

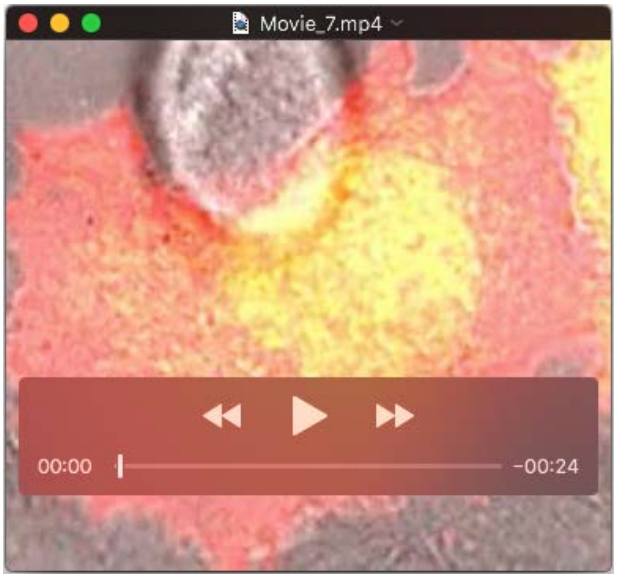
Movie 4. Time-lapse of HeLa cells transfected with Lifeact-RFP treated with CIT-K siRNA.



Movie 5. Time-lapse of HeLa cells transfected with Lifeact-RFP expressing GFP.



Movie 6. Time-lapse of HeLa cells transfected with Lifeact-RFP expressing CITK-GFP.



Movie 7. Time-lapse of HeLa cells transfected with Lifeact-RFP expressing ANLN-GFP.

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