

Figure S1. Diverse signaling molecules appear enriched in cortical structures. (A-D) HiLo microscopy images of mCherry::PH_{PLC δ 1} (n>5) (A), GFP::RHO-1 (n = 2) (B), CDC-42::mCherry (n = 3) (C), and GFP::CSNK-1 (n = 2) (D) at the cell cortex are shown from early symmetry-breaking to maintenance phase. All show cortical structures with a distinct pattern of appearance, subsequent enrichment in the anterior and dissipation upon entry to maintenance phase. Time (s) relative to the end of establishment phase. Scale bar = 10 um.

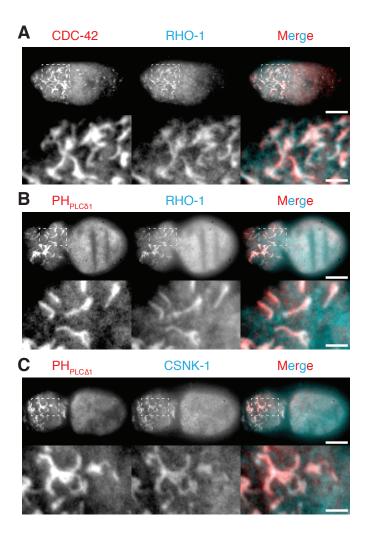


Figure S2. Colocalization of signaling molecules with the PIP₂ probe PH_{PLC δ 1}. (A) Sample image of surface of embryo co-expressing GFP::RHO-1 and mCherry CDC-42. Individual channels and merge shown with zoom of inset region to highlight structures. (B) As in (A), but GFP::RHO-1 with mCherry::PH_{PLC δ 1}. (C) As in (A), but GFP::CSNK-1 with mCherry::PH_{PLC δ 1}. Co-enrichment quantified in Figure 1B. Scale bars = 10 um.

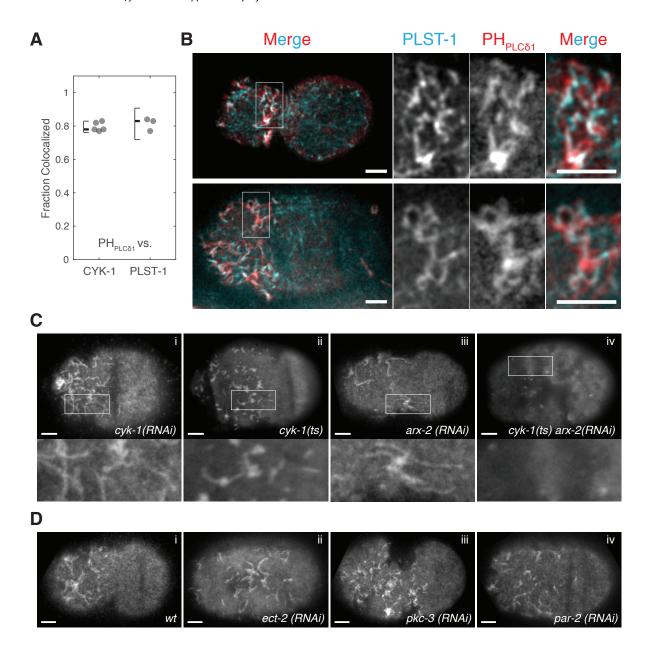


Figure S3. Filopodia are enriched in actin bundling proteins, require actin polymerization, and respond to polarity cues. (A) Fraction of selected $PH_{PLC\delta1}$ -labeled structures that exhibit colocalized CYK-1 puncta at tips or enrichment in PLST-1. Datapoints from individual embryos shown. Median \pm 95%CI indicated. (B) Cortical images of two embryos expressing PLST-1::GFP and mCherry::PH_{PLC\delta1}. Inset is marked and magnified on right showing merged and single channel images. (C) Disruption of both CYK-1 and ARX-2 lead to loss of filopodia like structures. mCherry (i,iii) or GFP (ii,iv) fusions to PH_{PLC\delta1} persist when either ARX-2 or CYK-1 are disrupted on their own, but not when both are compromised. Insets magnified below to highlight filopodia structure. (D) Formation of filopodia is not affected when contractility is compromised by depletion of the RhoGEF, ECT-2, or when polarity is disrupted by depletion of PKC-3 or PAR-2 by RNAi. However, asymmetry of filopodia is reduced in all three cases consistent with their asymmetry requiring polarity. ECT-2 depletion compromises symmetry-breaking during the establishment phase (*Zonies et al., 2010*). Images shown reflect a timepoint of peak filopodial density just before relaxation of the cortex and filopodial disassembly. Scale bars = 5 μ m.

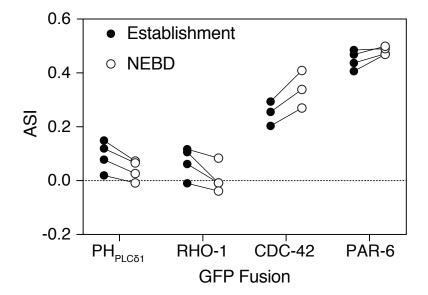


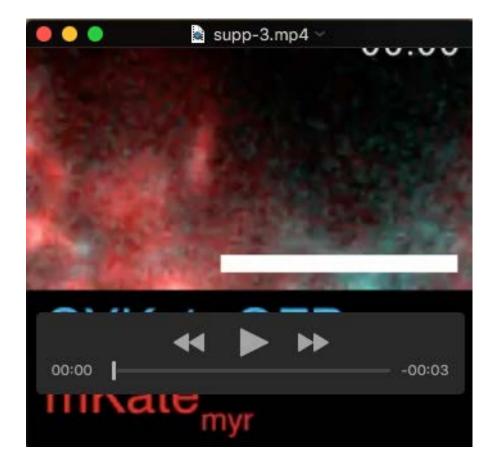
Figure S4. Asymmetry in membrane concentration from midplane images. Membrane fluorescence profiles were traced from midplane images of embryos expressing the indicated GFP fusions at mid-establishment (5 min pre-NEBD) or mid-maintenance phase (NEBD). For each embryo the asymmetry index (ASI) was calculated at each timepoint from the extracted profiles. Note asymmetry of both $PH_{PLC\delta1}$ and RHO-1 is low at establishment phase and is reduced further at maintenance phase when filopodia dissipate, consistent with filopodia contributing to weak apparent asymmetry during the establishment phase. By contrast, CDC-42 exhibits moderate asymmetry during establishment when anterior filopodia are high, but its asymmetry increases further by NEBD, when filopodia dissipate. For reference, the polarity protein PAR-6, which is not prominently enriched in these structures due to association with PAR-3 clusters, is highly polarized at both time points. Together these data suggest that filopodia-like structures, despite being highly asymmetric, do not contribute significantly to overall polarity in membrane concentrations.

 Table S1. C. elegans strains used in this work.

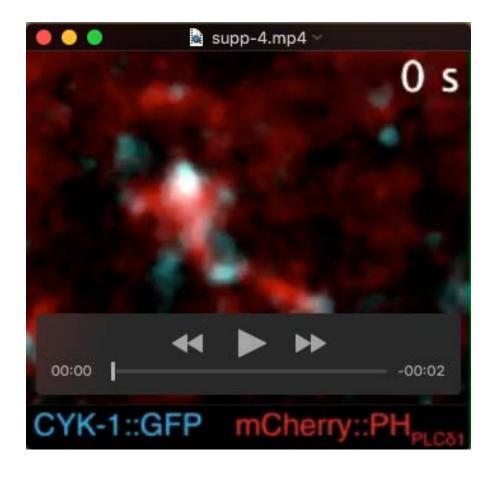
Strain	Genotype	Source
ACR004	cyk-1(ges1[cyk-1::eGFP + LoxP unc-119(+) LoxP] III; unc-119 (ed3) III;	This work
	ltls44pAA173; [pie-1p::mCherry::PH(PLC1delta1) + unc-119(+)] V	
ACR010	plst-1(ges2[plst-1::eGFP + LoxP unc-119(+) LoxP] IV; unc-119 (ed3) III;	This work
	ltls44pAA173; [pie-1p::mCherry::PH(PLCδ1) + unc-119(+)] V	
AD189	unc-119(ed3)	CGC, Kadandale et al. (2005)
FT204	unc-119(ed3)	CGC
JA1354	unc-119(e2498)	CGC, Panbianco et al. (2008)
JCC146	cyk-1(or596ts); unc-119(ed3); ltls38 [pAA1; pie-1p::GFP::PH(PLC δ 1) + unc-119	Jordan et al. (2016)
	(+)]; ltls37 [pAA64; pie-1p::mCherry::his-58; unc-119 (+)]IV	
KK1248	par-6(it310[par-6::gfp])	CGC, Ken Kemphues
NWG0045	unc-119(ed3)	This work
NWG0047	unc-119(ed3)III; crkEx1[pNG19: mex-5p::PH(PLCδ1)::GBP::mKate::nmy- 2 3′UTR	Rodriguez et al. (2017)
	+ unc-119(+)]; him-5 (e1490) V	
OD58	unc-119(ed3) III; ltls38[pAA1; pie-1p::GFP::PH(PLCδ1) + unc-119(+)]	CGC, Audhya et al. (2005)
OD70	unc-119(ed3) III; ltls44pAA173; [pie-1p-mCherry::PH(PLCδ1) + unc-119(+)] V	CGC, <i>Kachur et al.</i> (2008)
SA115	tjls1[pie-1::GFP::rho-1 + unc-119(+)]	CGC, Motegi and Sugimoto (2006
SWG1	mex-5p::Lifeact::mKate2	Reymann et al. (2016)
SWG5	plst-1(ges2[plst-1::eGFP + LoxP unc-119 (+) LoxP] IV; unc-119 (ed3) III)	Reymann et al. (2016)
SWG19	cyk-1(ges1[cyk-1::eGFP + LoxP unc-119(+) LoxP) III; unc-119(ed3) III.	Reymann et al. (2016)
TH159	ddls46[WRM0625bA11 GLCherry::cdc-42; Cbr-unc-119(+)]	Rodriguez et al. (2017)
TH220	unc-119(ed3) III; ddls86[pie-1p::LifeAct::GFP; unc-119(+)]	Redemann et al. (2010)
WS5018	cdc-42(gk388) opls295[cdc-42p::GFP::cdc-42(genomic)::cdc-42 3'UTR + unc-	CGC, Neukomm et al. (2014)
	119(+)] II.	



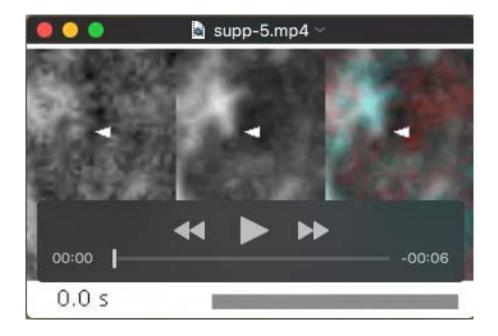
Movie 1. Timelapse video of the surface of a permeabilized embryo expressing $PH_{PLC\delta 1}$::GFP (middle, cyan) and stained with FM4-64 (left, red). Scale bar, 2.5 μ m. Elapsed time (sec). See related Figure 1F.



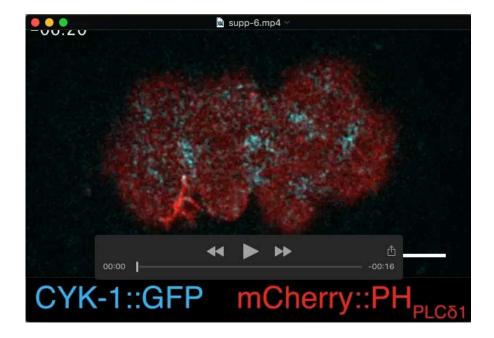
Movie 2. Timelapse video of filopodia extending into the pseudocleavage furrow in an embryo expressing CYK-1::GFP and mKate_{myr}. Scale bar, 5 μ m. Elapsed time (mm:ss) shown. See related Figure 2A.



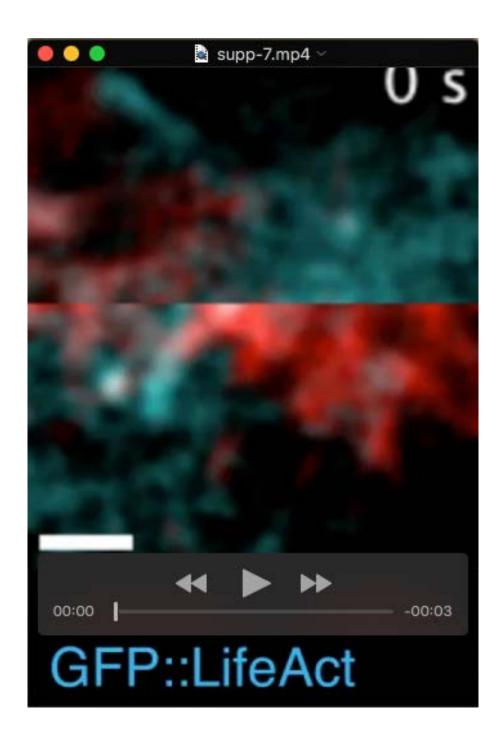
Movie 3. Timelapse video of dynamic filopodia on the surface of an embryo expressing CYK-1::GFP and $_{PLC\delta 1}$::mCherry. Scale bar, 5 μ m. Elapsed time (sec) shown. See related Figure 2D.



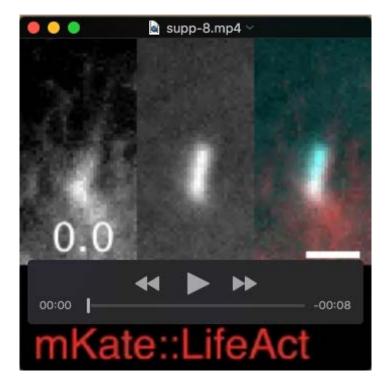
Movie 4. Timelapse video of an extending filopodium labeled with LifeAct::GFP and $_{PLC\delta 1}$::mCherry on the surface of a one-cell embryo. Elapsed time (sec) shown. Scale bar, 5 μ m. See related Figure 2F.



Movie 5. Timelapse video of the surface of an embryo expressing CYK-1::GFP and $PH_{PLC\delta 1}$::mCherry by confocal microscopy showing the appearance, accumulation, segregation and dissipation of filopodia. Elapsed time shown (mm:ss). Scale bar, 5 μ m. See related Figure 2K.



Movie 6. Timelapse video of two filopodia labeled with LifeAct::mKate (red) and LifeAct::GFP (cyan) corresponding to straightened filopodia shown in Figure 3E. Note lag of LifeAct::mKate signal relative to LifeAct::GFP yielding a cyan tip followed by a red tail. Elapsed time shown (sec). Scale bar, 2.5 µm. See related Figure 3F-G.



Movie 7. Timelapse video of cytoplasmic actin comet labeled with LifeAct::mKate (red) and LifeAct::GFP (cyan). Note lag of LifeAct::mKate signal relative to LifeAct::GFP yielding a cyan tip followed by a red tail. Elapsed time shown (sec). Scale bar, 2.5 μm. See related Figure 3G-H.

References

- Audhya A., Hyndman F., McLeod I.X., Maddox A.S., Yates J.R., Desai A., Oegema K. (2005). A complex containing the Sm protein CAR-1 and the RNA helicase CGH-1 is required for embryonic cytokinesis in *Caenorhabditis elegans*. *J. Cell Biol.* 171, 267–279.
- **Cuenca A.A., Schetter A., Aceto D., Kemphues K., Seydoux G.** (2003). Polarization of the *C. elegans* zygote proceeds via distinct establishment and maintenance phases. *Development* 130, 1255–1265.
- **Jordan S.N., Davies T., Zhuravlev Y., Dumont J., Shirasu-Hiza M., Canman J.C.** (2016). Cortical PAR polarity proteins promote robust cytokinesis during asymmetric cell division. *J. Cell Biol.* 212, 39–49.
- **Kachur T.M., Audhya A., Pilgrim D.B.** (2008). UNC-45 is required for NMY-2 contractile function in early embryonic polarity establishment and germline cellularization in *C. elegans. Dev. Biol.* 314, 287–299.
- **Neukomm L.J., Zeng S., Frei A.P., Huegli P.A., Hengartner M.O.** (2014). Small GTPase CDC-42 promotes apoptotic cell corpse clearance in response to PAT-2 and CED-1 in *C. elegans. Cell Death Differ.* 21, 845–853.
- Redemann S., Pecreaux J., Goehring N.W., Khairy K., Stelzer E.H.K., Hyman A.A., Howard J. (2010). Membrane Invaginations Reveal Cortical Sites that Pull on Mitotic Spindles in One-Cell *C. elegans* Embryos. *PLoS ONE*. 5, e12301.
- **Zonies S., Motegi F., Hao Y., Seydoux G.** (2010). Symmetry breaking and polarization of the *C. elegans* zygote by the polarity protein PAR-2. *Development* 137, 1669–1677.