



Figure S1. Morphological changes with RASSF4

A. Cell height determined by z-stack analysis for control cells overexpressing CFP-PH-PLC δ 1 and those co-expressing GFP-PIP5KI β , GFP-PIP5KI γ , or RASSF4-YFP. **B.** Capacitance of control cells and those overexpressing GFP-PIP5KI β , GFP-PIP5KI γ , or RASSF4-YFP. Cells were patched in whole-cell voltage-clamp configuration with 50-60% compensation of series resistance. **C.** Airyscan confocal images of CFP-PH-PLC δ 1 in control cells (left) and cells co-expressing RASSF4-YFP. Images were taken at the glass-adhered basal membrane of the cells and yellow lines determined by thresholding delineate filopodia and lamellipodia. Increased fluorescent intensity in this region corresponds to two membrane surfaces in the image plane. **D.** Percentage of cross-sectional area (from C) occupied by filopodia and lamellipodia in cells labeled with CFP-PH-PLC δ 1. **E.** Percentage of cross-sectional area occupied by filopodia and lamellipodia in cells labeled with Tubby_C^{R332H}. **F.** Airyscan confocal images for cells expressing Tubby_C^{R332H} alone (left) or with co-expression of RASSF4-YFP (right). Four z-stack images of a single cell are collapsed to highlight the presence of intracellular bodies. **G.** The number of Tubby_C^{R332H}-labeled intracellular bodies in control cells and those co-expressing RASSF4. **H.** Total fluorescence of Tubby_C^{R332H}-labeled intracellular bodies in control cells and those co-expressing RASSF4. **I.** Representative mid-section confocal micrographs from cells expressing RASSF4-mCh with or without GFP-PIP5KI β . Contrast is inverted to show RASSF4-mCh fluorescence as black. * $p \leq 0.05$; *** $p \leq 0.0001$ compared to control cells by Student's t-test (one-tailed).