Supplemental Movie 1. Rapamycin-induced inhibition of the capsaicin current tracks depletion of $PI(4,5)P_2$ in the plasma membrane. Whole-cell current measured using perforated-patch configuration (upper panel) and simultaneous confocal imaging of the fluorescent $PI(4,5)P_2$ probe, YFP-PH-PLCδ1 (see Experimental Procedures). Same cell as in Figure 4. Frame interval, 8 s.

Supplemental Movie 2. Rapamycin induced translocation of CFP-FKBP-Ins-5P to the plasma membrane and depletion of $PI(4,5)P_2$. Confocal images of two adjacent cells, both of which have been transfected with all the components of the rapamycin-inducible Ins-5P system and YFP-PLCδ1-PH probe. Images were obtained with a Zeiss LSM 510 META confocal microscope using a 63X-water immersion objective (Keck Imaging Center, University of Washington). The CFP channel (excitation 458 nm, emission 470-500 nm) is shown on the left and the YFP channel (excitation 514 nm, emission 530-600) is shown on the right, as indicated. Images were acquired sequentially to avoid fluorescence bleed through into the wrong acquisition channel. Our bath solution was a Hank's Buffered Salt Solution (in mM: 140 NaCl, 4 KCl, 1 $MgCl_2$, 1.8 $CaCl_2$, 5 glucose, 10 HEPES, pH 7.4). Application of rapamycin (1 μM) caused translocation of CFP-FKBP-Ins-5P from the cytosol to the plasma membrane. This translocation was followed by a translocation of YFP-PLCδ1-PH from the plasma membrane to the cytosol, indicating that plasma membrane $PI(4,5)P_2$ was depleted. Frame interval, 10 s.