Supplementary Material

Figure S1: Selective Labeling of Subcellular compartments in COS-7 cells. Projection images of confocal stacks shown. **A)** Representative surface membrane staining using CT-B-594 at 4 C. **B)** Labeling of ER-golgi using rBrefA staining. +Vis - overlay of DIC image shows that cell margin extends well beyond the bright perinuclear staining produced by rBrefA. **C)** Characteristic fluorescence image of a COS-7 cell expressing a small soluble EGFP protein that fills the entire cytoplasm, including the nucleus. Scale bars are 20 μm.

Figure S2: Subcellular distribution of wild type subunits co-expressed in COS7 cells.

Channel constructs expressed in COS7 cells. KChIP3 and DPP6 detected by immunofluorescence. Kv4.2(EGFP) detected by green fluorescence. Subcellular internal membranes (rBrefA) and surface membrane (CT-B-594) imaged with red fluorescence. Projection images of confocal stacks shown. **A**) KChIP3 shows a subcellular distribution expected for a soluble protein, similar to EGFP (see Fig. S1C). **B**) DPP6 expressed alone targets membrane compartments with considerable protein on the surface membrane. **C**) Distribution of Kv4.2(EGFP) expressed alone compared to rBrefA, shows the channel strongly co-localizes with internal membranes. Visible DIC image shows the cell margin extends well beyond the perinuclear channel fluorescence. **D**) Kv4.2(EGFP) expressed alone shows no co-localization with the surface membrane marker CT-B-594. **E**) Kv4.2(EGFP) co-expressed with KChIP3 redistributes to strongly co-localize with the surface membrane marker CT-B-594. **F**) Kv4.2(EGFP) co-expressed with DPP6 also redistributes to strongly co-localize with the surface membrane marker CT-B-594. Scale bars are 20 μm.



Figure S1

