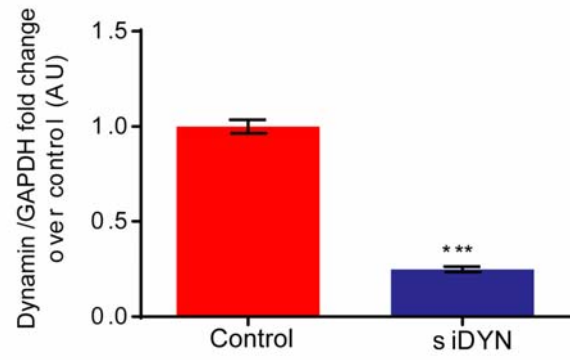
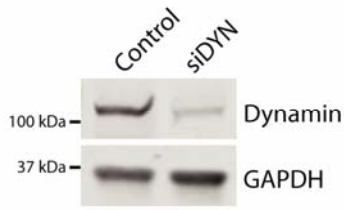


Supplemental Materials

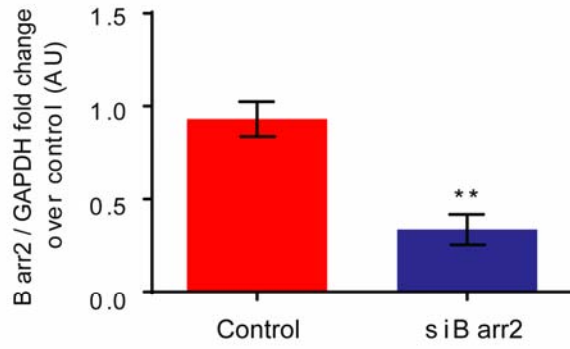
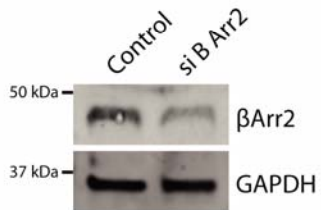
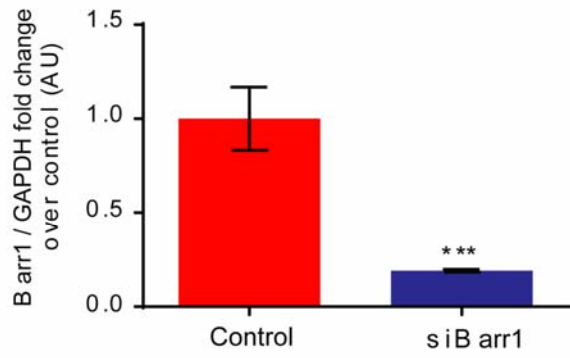
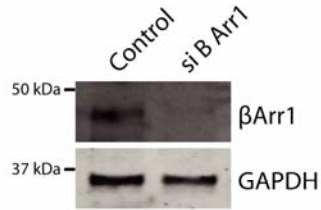
Molecular Biology of the Cell

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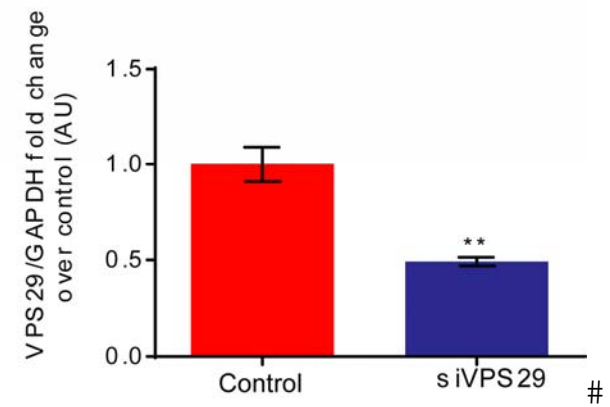
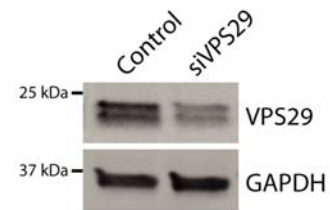
A



B



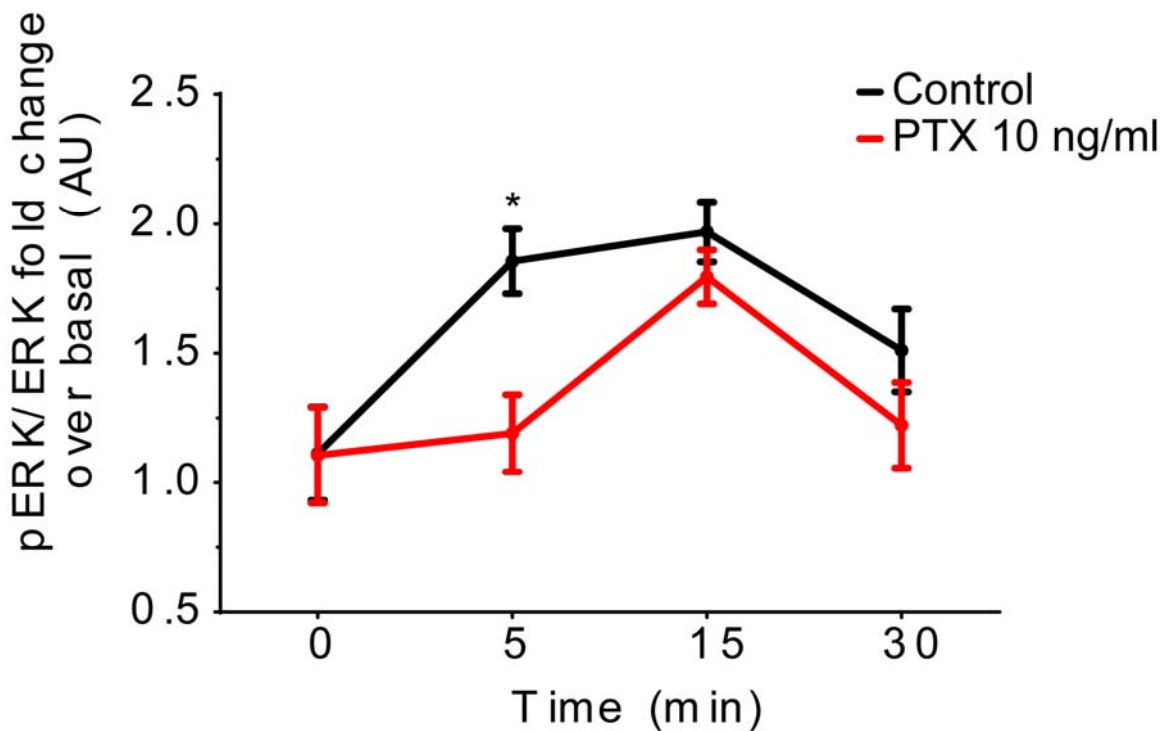
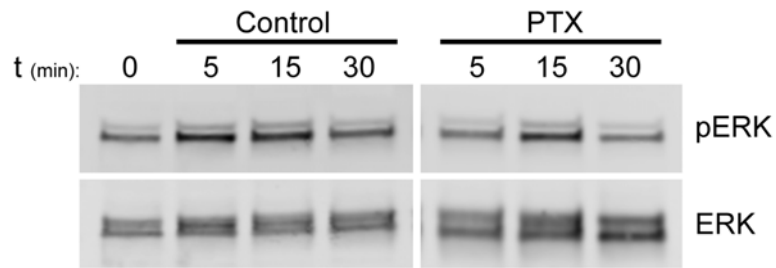
C



Supplemental figure 1

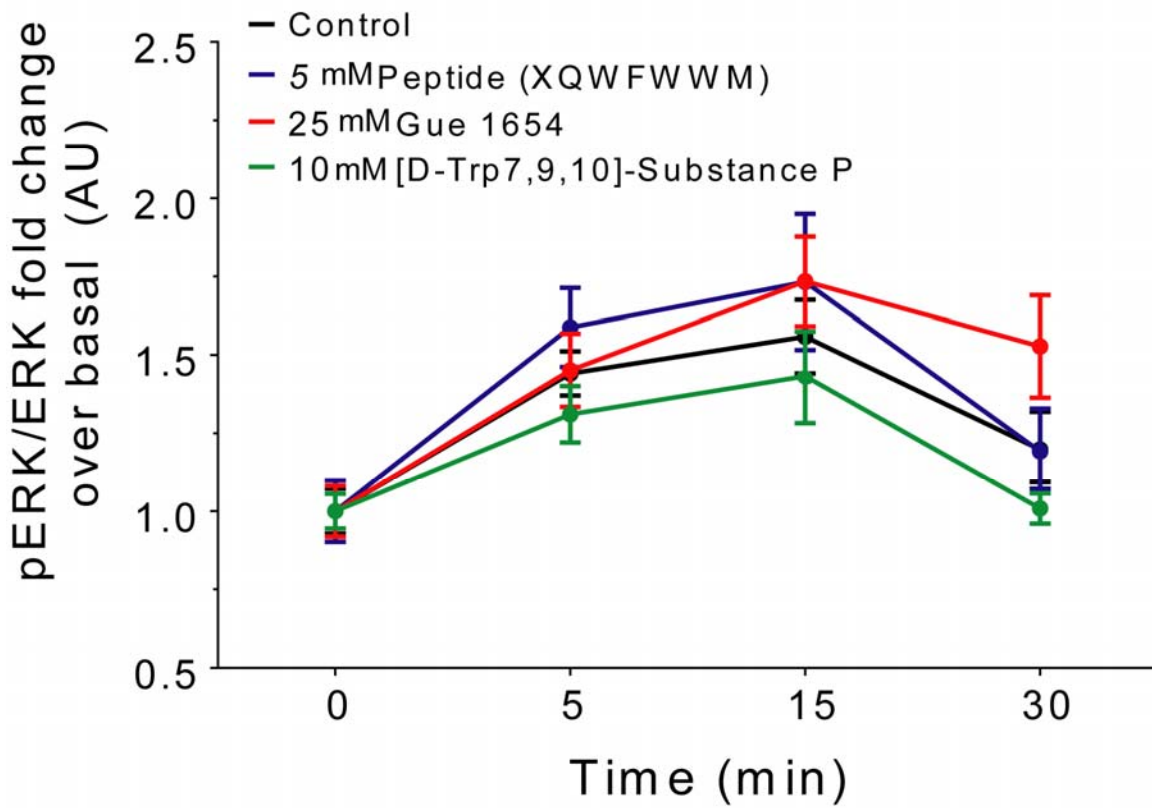
The effectiveness of siRNA knockdown was confirmed by western-blot. (A) Dynamin expression levels was investigated two to three days after siRNA transfection utilizing Dynamin 2 specific antibody. Significant reduction in protein expression was observed in multiple experiments (B) siRNA knockdown of beta-arrestin1 and beta arrestin 2 was confirmed with specific antibodies. (C) Reduction of VPS29 expression levels by siRNA was analyzed by western-blot. Quantifications were performed as fold change over control (GAPDH), n>5.

HEK-CB1R



Supplemental figure 2

(A) Time course of ERK1/2 phosphorylation by 2-AG in HEK cells stably expressing CB₁R was analyzed by western-blot. Cell lysates were analyzed with phospho-ERK (top) and total ERK (bottom panel). Cells were preincubated with 10ng/ml PTX overnight, before 2-AG treatments. (B) Time course quantification of independent experiments (n=5)



Supplemental figure 3

Time course showing ERK1/2 phosphorylation in HEK293 cells stably expressing CB₂R and preincubated with G protein inhibitors as describe in methods in the presence of 10 μ M 2-AG. Cell lysates were analyzed by western blots against phospho-ERK1/2 and total ERK1/2 and quantified (n>6). No statistical significance was observed between treatments and control samples.