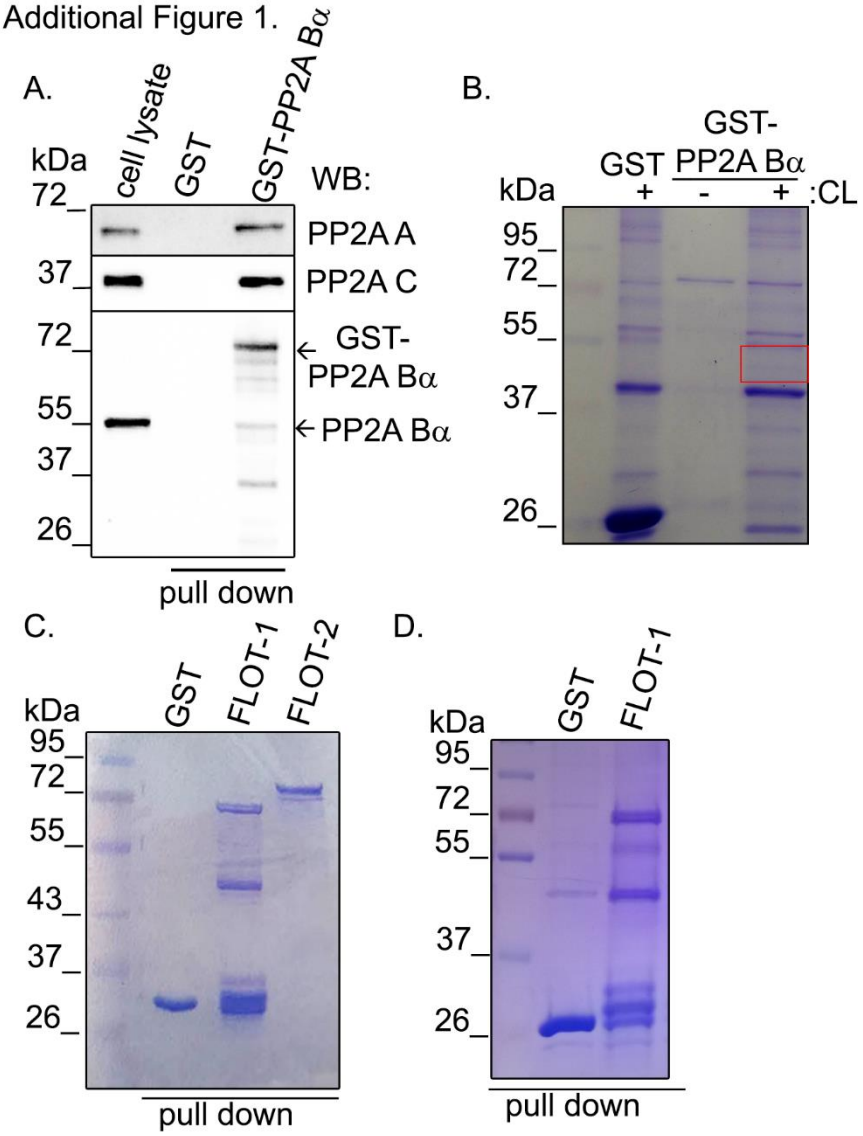
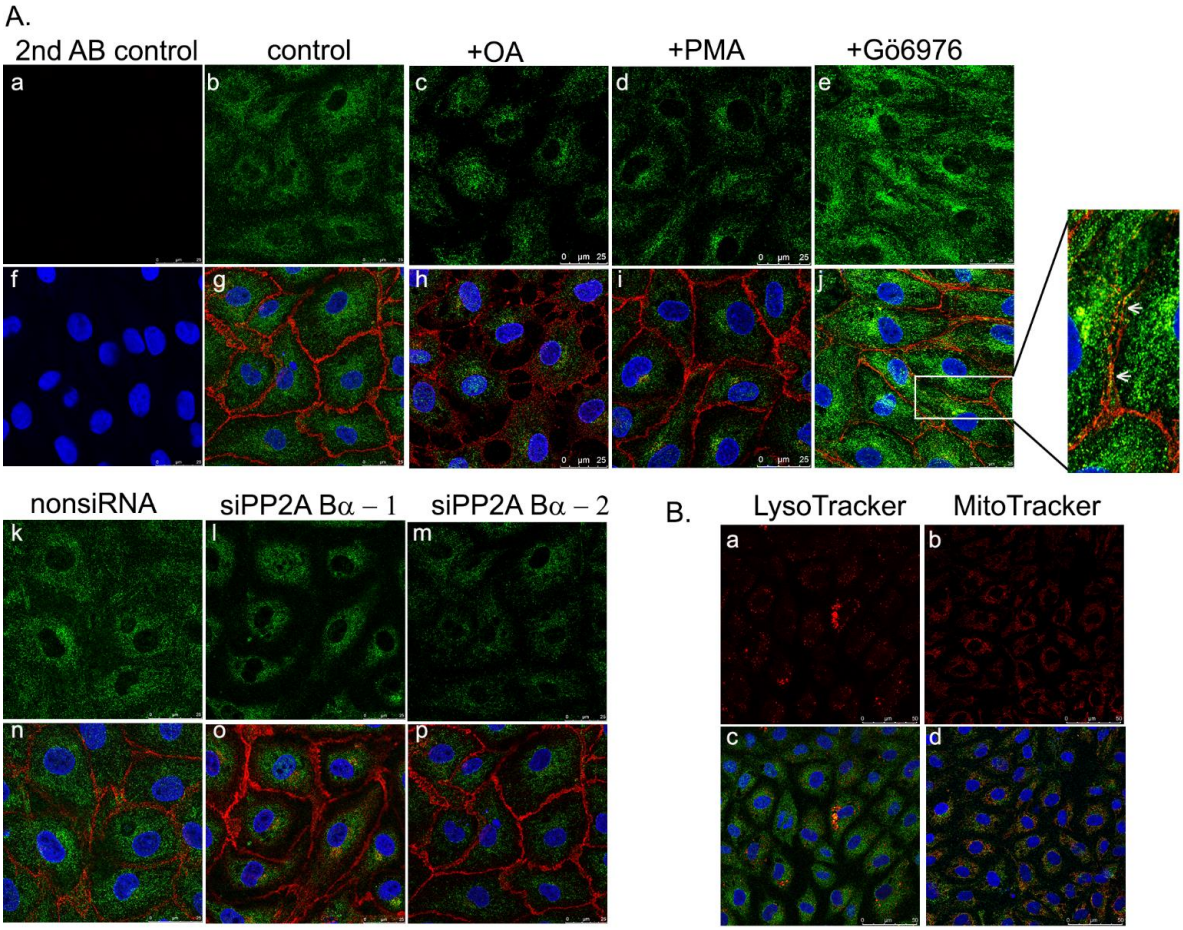
Additional Figure 1.



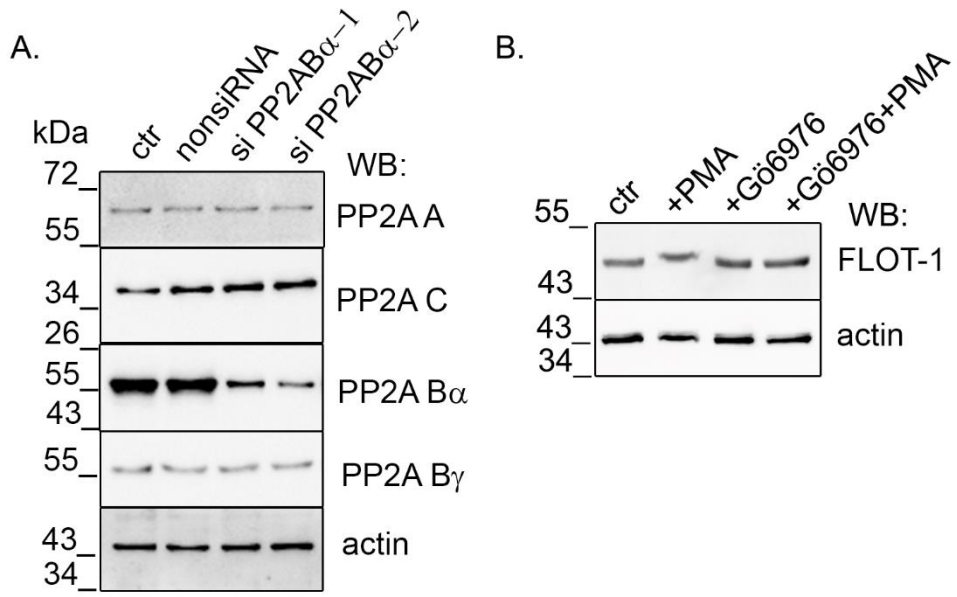
Additional figure 1. Internal controls of the interaction between PP2A Bα and flotillin proteins A) Western blot analysis of endothelial cell lysate and pull down samples of GST and GST- PP2A Bα. B) Bacterially expressed GST and GST-tagged PP2A Bα were loaded onto glutathione-Sepharose. After washing steps the resin samples were incubated with BPAEC lysate (CL) or cell lysis buffer . The bound proteins were eluted by boiling the samples in 2x SDS sample buffer. Blue silver staining of the eluted fractions after the pull-down are shown. Red box shows the band of a possible interacting protein appearing only in the GST-tagged PP2A Bα sample incubated with EC cell lysate. That band was cut from the gel and was identified by LC-MS/MS as flotillin-1 which was confirmed by Western blot (Fig 1A). C-D) Coomassie Blue staining of the pull down samples (corresponding to Fig 1B and Fig 1D, respectively) shows the efficiency of purification and the presence of GST-tagged recombinant proteins.

Additional Figure 2.



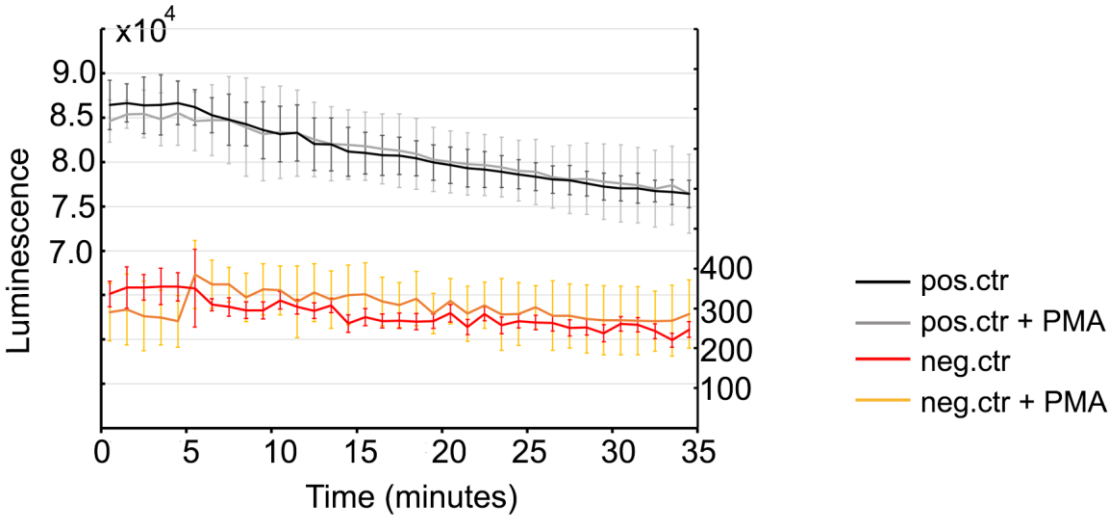
Additional figure 2. Localization of flotillin-1 in endothelial cells. A) Immunfluorescence staining of BPAEC using flotillin-1 (green) and VE-cadherin (red). Zoomed section (marked by white rectangle) on merged image "j" show flotillin-1 appearance in membrane region. B) Lysosome specific LysoTracker dye (red), mitochondria specific MitoTracker dye (red) staining of BPAEC is shown. Flotillin-1 was stained with anti-flotillin-1 specific antibody (green) and DAPI (blue) was used to visualize nuclei. Scale bar: 50µm.

Additional Fig.3



Additional figure 3. Efficiency of PP2A Bα silencing. A) Control, non siRNA and PP2A Bα specific siRNA transfected BPAEC cells were studied by Western blot using anti-PP2A A, anti-PP2A C, anti-PP2A Bα, anti-PP2A Bγ and actin specific antibodies. B) BPAEC cells were treated with PMA, Gö6976 and PMA after addition of Gö6976. Proteins were resolved on 9% SDS-PAGE using 60V to detect mobility shift of flotillin-1. Samples were analysed by Western blot using flotillin-1 and actin antibody. Flotillin-1 molecular weight was calculated using Image Lab software.

Additional Fig. 4.



Additional figure 4. Controls of NanoBiT luciferase complementation assay. BPAEC cells co-transfected with SmBiT-PRKACA and LgBiT-PRKAR2A were used as positive control. Negative control vectors SmBiT-Halotag and LgBiT-PP2A Bα were also used.

Graphical abstract