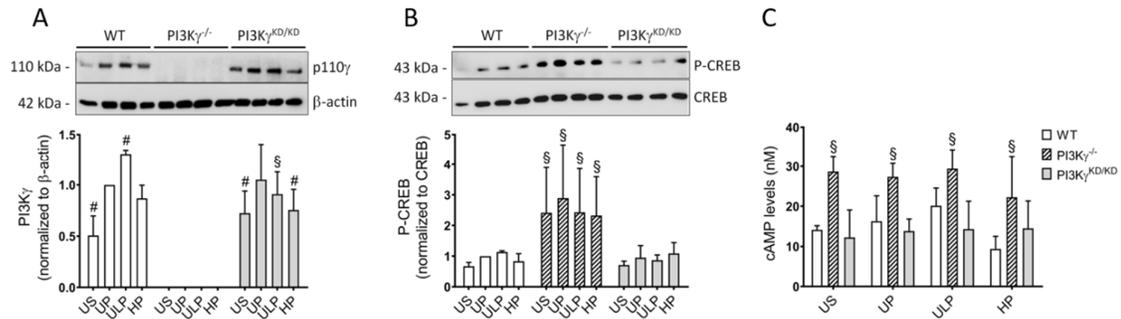
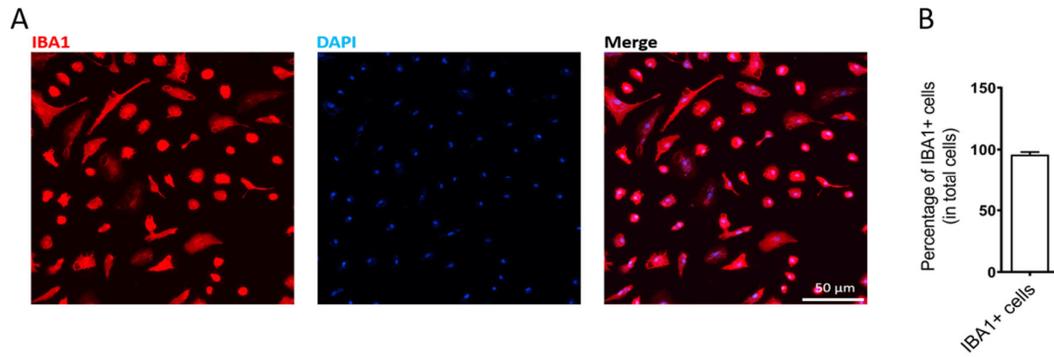


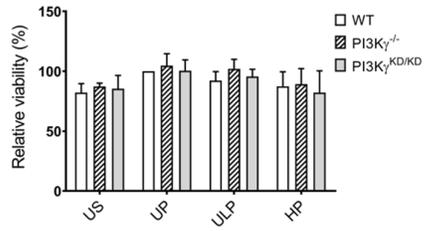
Supplementary Figures



Supplementary Figure 1. Impact of microglia stimulation by LPS including dose-dependent priming of PI3K γ expression and effects of lipid kinase dependent and independent PI3K γ signaling on parameters of intracellular signaling. Primary microglial cells (wild-type, open columns; PI3K $\gamma^{-/-}$, hatched columns; PI3K $\gamma^{KD/KD}$, dark gray columns) were primed initially by ultra-low (ULP, 1 fg/mL) or high (HP, 100 ng/mL) doses of LPS, followed by a second stimulation (day 6) with 100 ng/mL LPS. Lysates were collected 24h after the second stimulus and analyzed for (A) p110 γ protein expression (n=6) and (B) phospho-CREB protein expression (n=5) using Western blotting and quantified (wild-type unprimed cells assigned as 1.0). (C) cAMP levels (n=5) were assayed using the cAMP GloAssayKit. Data are shown as mean + SD, [§]p < 0.05, [#] vs. unprimed condition, [§] vs. wild-type strain. US, unstimulated; UP, unprimed.



Supplementary Figure 2. Purity of primary microglial cell culture assessed by the percentage of IBA1-positive cells (A, red color) in total cells indicated by immunocytochemical staining using DAPI (blue color). N=6, at least 5 randomly selected fields were used for quantification (B). Data are shown as mean + SD.



Supplementary Figure 3. Cell viability in primary microglial cell culture primed by LPS.

Primary microglial cells (wild-type, open columns; PI3K $\gamma^{-/-}$, hatched columns; PI3K $\gamma^{KD/KD}$, dark gray columns) were primed initially by ultra-low (ULP, 1 fg/mL) or high (HP, 100 ng/ml) doses of LPS, followed by a second stimulation (day 6) with 100 ng/ml LPS. Cell viability was measured using MTT assay (n=4, wild-type cells assigned as 100%). Data are shown as mean + SD, # § p < 0.05, # vs. unprimed condition of wild-type microglia, § vs. wild-type strain. US, unstimulated; UP, unprimed.