

SUPPLEMENTAL INFORMATION:

Activity and Stability of Panx1 Channels in Astrocytes and Neuroblastoma Cells Are Enhanced by Cholesterol Depletion

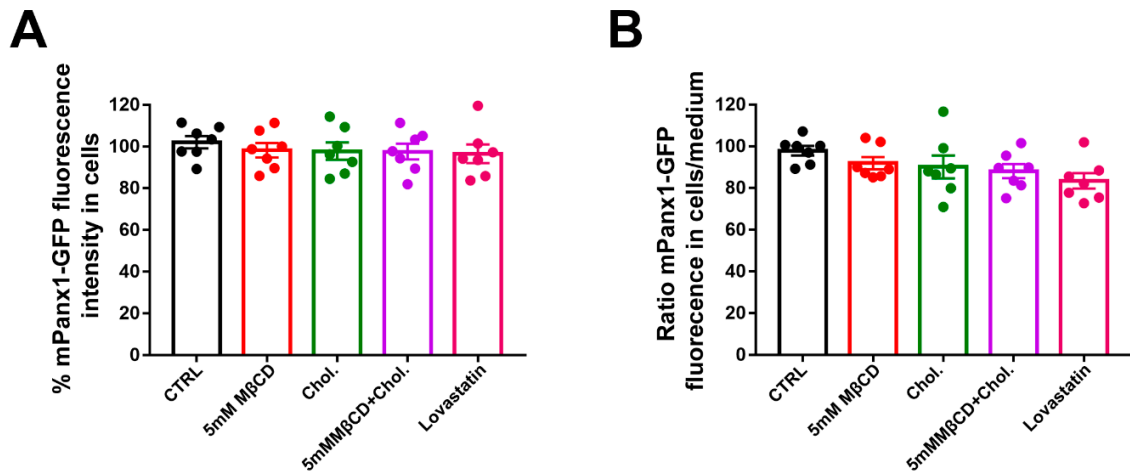
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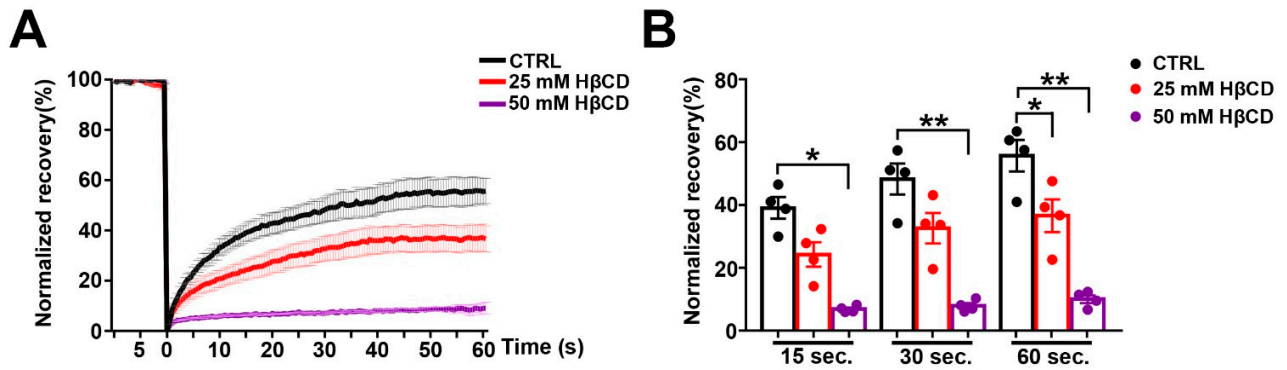
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Supplemental Figure S1. Effects of treatment with MβCD and lovastatin on mPanx-1-GFP expression.

mPanx-1-GFP transfected N2a cells were incubated with cyclodextrin (5 mM MβCD for 60 min at 37°C), lovastatin (5 μM, 12 hr) in serum-free DMEM, cholesterol (2h, 37°C) or left untreated (CTRL). (A) Histograms showing mPanx-1-GFP fluorescence intensity in cells expressed as a percentage relative to that of untreated cells (CTRL), considered as 100 %. Note that treatments did not effect mPanx1-GFP expression intensity in cells. Each point on the graphs represents relative mPanx1-GFP fluorescence changes recorded from all cells present in a field of view obtained from at 3 independent experiments. (B) Ratios of fluorescence intensity of mPanx-1-GFP in cells to medium. Note that the ratios of intensity are similar. ANOVA test followed by Dunnett's test.



Supplemental Figure S2. Effects of 2-Hydroxypropyl- β -cyclodextrin on the lateral diffusion of mPanx-1-GFP. N2a cells transiently expressing mPanx-1-GFP were subjected to treatments with 25 mM and 50 mM of 2-Hydroxypropyl- β -cyclodextrin (H β CD) or left untreated (CTRL). FRAP experiments were conducted on live cells at 30°C. (A) FRAP recovery curves for control cells and with 25 mM and 50 mM of H β CD. (B) Histograms of normalized FRAP recovery at 15, 30 and 60 sec after photobleach. $n=7-10$ per plasma membrane domain, data collected over three independent repeats. P values obtained from ANOVA test followed by Dunnett's test. (*: $p<.05$; **: $p<.01$).