



ESM Fig. 1 NPY-pHluorin visualizes the fraction of released insulin granules. **a** Confocal image of an intact human islet infected with NPY-pHluorin taken 90 s after stimulation with NH₄Cl (50 mmol/l). A pseudocolor scale was used to illustrate better the changes in fluorescence intensity. Scale bar, 10 μ m. **b, c** Zoomed confocal images of cells highlighted with boxes in **a**. Images were taken under basal glucose (3 mmol/l, 3G), 1 min after KCl (30 mmol/l) or NH₄Cl (50 mmol/l) application. Scale bar, 5 μ m (**b, c**). **d** Traces showing changes in the mean fluorescence intensity of different cells within the islet upon sequential stimulation with KCl and NH₄Cl. Black traces refer to individual cells and red trace to the whole islet. **e** Quantification of changes in mean fluorescence intensity (maximum amplitude – baseline) induced by KCl and NH₄Cl (n = 19 cells). KCl depolarization induces an increase in fluorescence that is only a small fraction of that stimulated by NH₄Cl (~5%). Results are shown as mean \pm SEM.