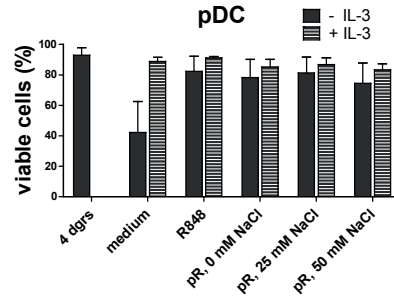
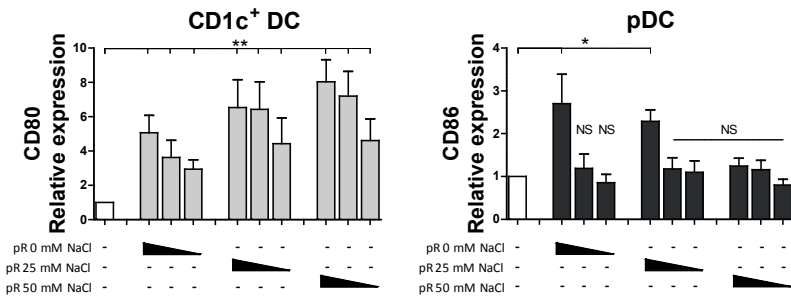


Supplementary figure 1

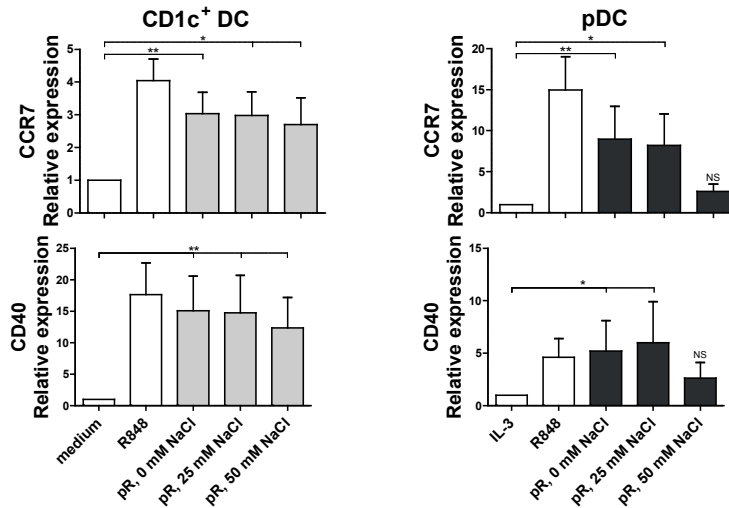
a



b



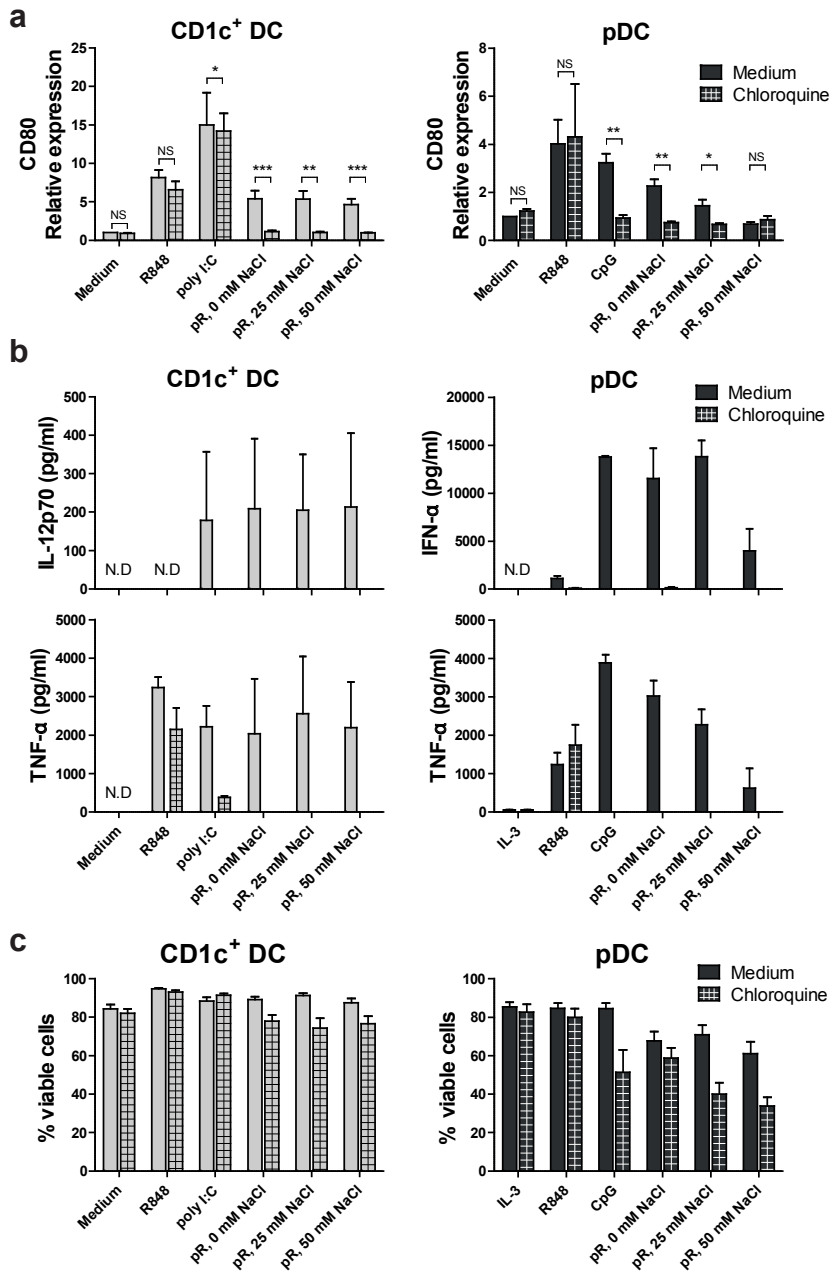
c



Supplementary figure 1: pDC viability in the presence or absence of IL-3 and DC maturation upon stimulation with protamine:RNA complexes

Purified CD1c⁺ DCs and pDCs were cultured 18-24h with 15 μ g/ml, 7.5 μ g/ml or 1.5 μ g/ml of protamine:RNA complexes (pR) formed in 0, 25, or 50 mM NaCl. (a) The viability of pDCs cultured with R848 or 15 μ g/ml of pR in the presence or absence of IL-3 was determined. The mean percentage \pm SEM of cells negative for live-dead marker from 4 pDC donors is depicted. (b) The relative expression of CD80 on DCs treated with medium or IL-3 or increasing concentration of pR is calculated by normalizing the MFI values for each donor against the negative control. The fold increase \pm SEM of 9-10 CD1c⁺ DC donors and 4-5 pDC donors is depicted. (c) The relative expression of CD40 and CCR7 from 7 CD1c⁺ DC and pDC donors treated with medium or IL-3, R848, or 15 μ g/ml of pR is depicted. Wilcoxon matched-pair signed rank tests were performed on raw data, comparing against negative control, and are indicated by * ($p < 0.05$), ** ($p < 0.01$), or NS (non-significant)

Supplementary figure 2



Supplementary figure 2: Activation and viability of chloroquine-treated DCs

CD1c⁺ DCs and pDCs were pre-incubated for 60 minutes with chloroquine before the addition of medium or IL-3, R848, poly I:C or CpG-C, or protamine:RNA complexes (pR) formed in 0, 25, or 50 mM NaCl. (a) The relative expression of CD80 on DCs was calculated by normalizing the MFI values for each donor against the negative control. Fold increase \pm SEM of 4-5 CD1c⁺ DC and pDC donors is depicted. Wilcoxon matched-pair signed rank tests were performed and are indicated by * ($p < 0.05$), ** ($p < 0.01$), *** ($p < 0.001$), or NS (non-significant). (b) The secretion of IL-12p70 from 3 CD1c⁺ DC donors, of IFN- α from 2-3 pDC donors, and of TNF- α from the same donors is depicted as mean percentage \pm SEM. N.D.=not detected. (c) The viability of DCs cultured with chloroquine is analyzed and the mean percentage \pm SEM of viable cells from 5 CD1c⁺ DC donors and 4 pDC donors is depicted.