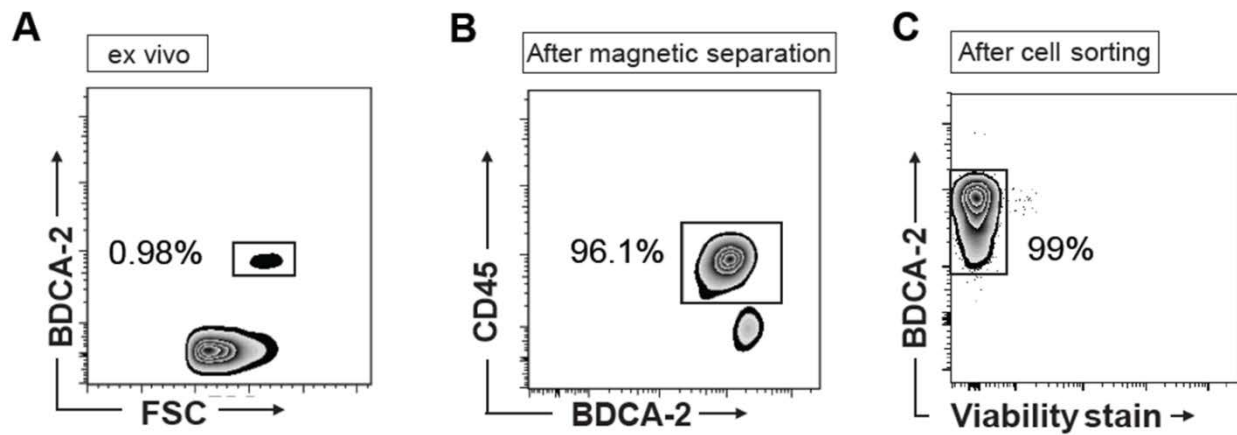
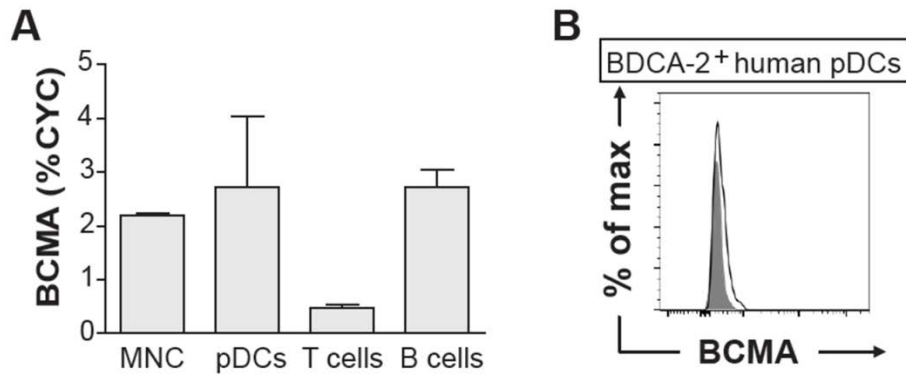


Suppl. Fig. 1



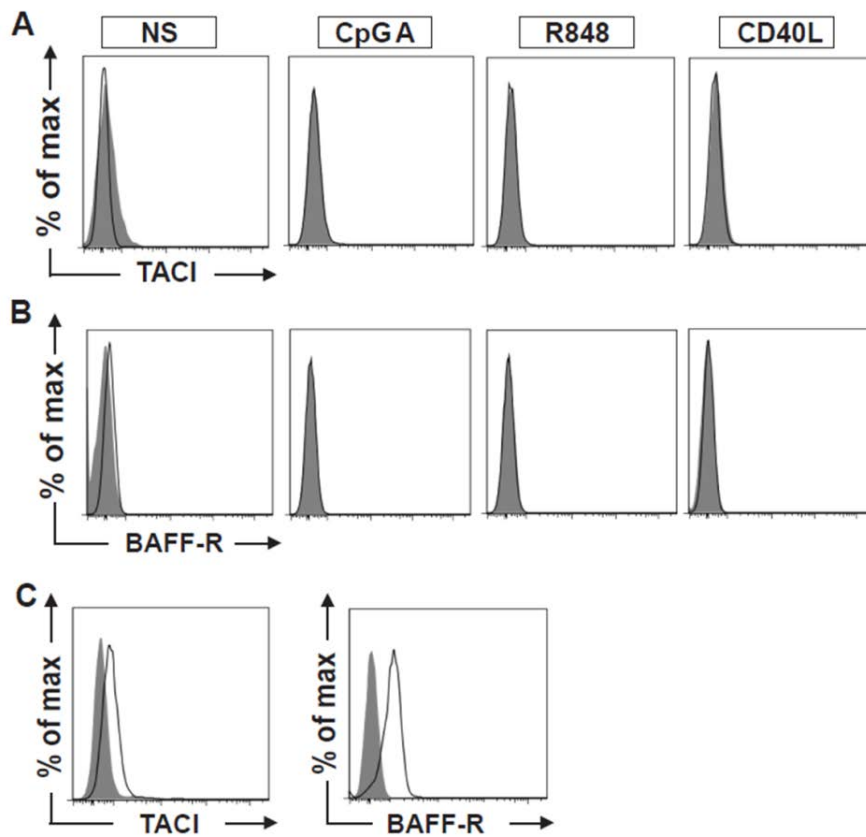
Supplementary figure 1. Stepwise isolation of primary human pDCs. To work with primary human pDCs we established a stepwise purification protocol of whole PBMCs. Each experiment represents one blood donor. **(A)** FACS staining of PBMCs. The frequency of pDCs in PBMCs ranged between 0.4-1%. **(B)** After a positive selection with magnetic beads and anti-BDCA-4, purity exceeded 95% as determined by BDCA-2⁺ and CD45⁺ surface expression. To enhance purity, BDCA-2⁺ cells were additionally sorted by FACS. **(C)** Purity levels exceeded 99% when gated on the living cell population.

Suppl. Fig. 2



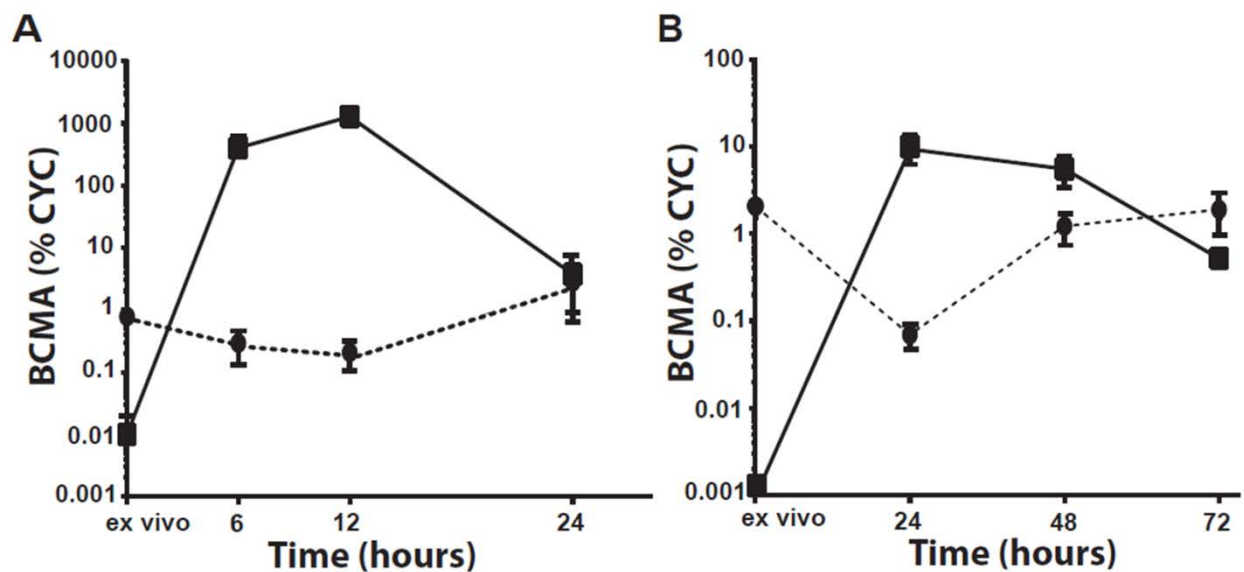
Supplementary figure 2. Tonsil derived pDCs contain BCMA transcripts, but lack BCMA protein on the cell surface. **(A)** The indicated immune cell subsets were purified with magnetic beads from tonsils and the expression levels of BCMA were determined by qPCR. Combined data of 3 independent experiments with 3 donors are depicted (mean \pm SEM). **(B)** Single cell suspensions from tonsils were analyzed by flow cytometry. The BDCA-2⁺ pDCs fraction shows a similar staining with anti-BCMA and control Ig. One representative of 3 independent experiments is shown. The closed histogram represents the isotype control, the solid black lines the BCMA surface expression.

Suppl. Fig. 3



Supplementary figure 3. Human pDCs do not express TACI and BAFF-R after activation. (A, B) pDCs purified from human blood were cultured for three days with CpG-A (ODN 2216), R848 or CD40L expressing mouse fibroblasts. Then, surface expression of BAFF-R and TACI was determined. (C) The Burkitt's lymphoma cell line RAJI was used as positive control for TACI and BAFF-R expression.

Suppl. Fig. 4



Supplementary figure 4. Transcript levels of BCMA and IFN- α 1 after stimulation of human pDCs with CpG-A. Human pDC were sorted from blood, stimulated with CpG-A and cultured for the indicated time points. Subsequently RNA was obtained, cDNA prepared and a qPCR was performed for BCMA (dotted line) and IFN- α 1 (solid line). Three experiments were performed to analyse the expression levels after a short time (A) and three others to examine this after a longer culture period (B). Error bars indicate SEM.