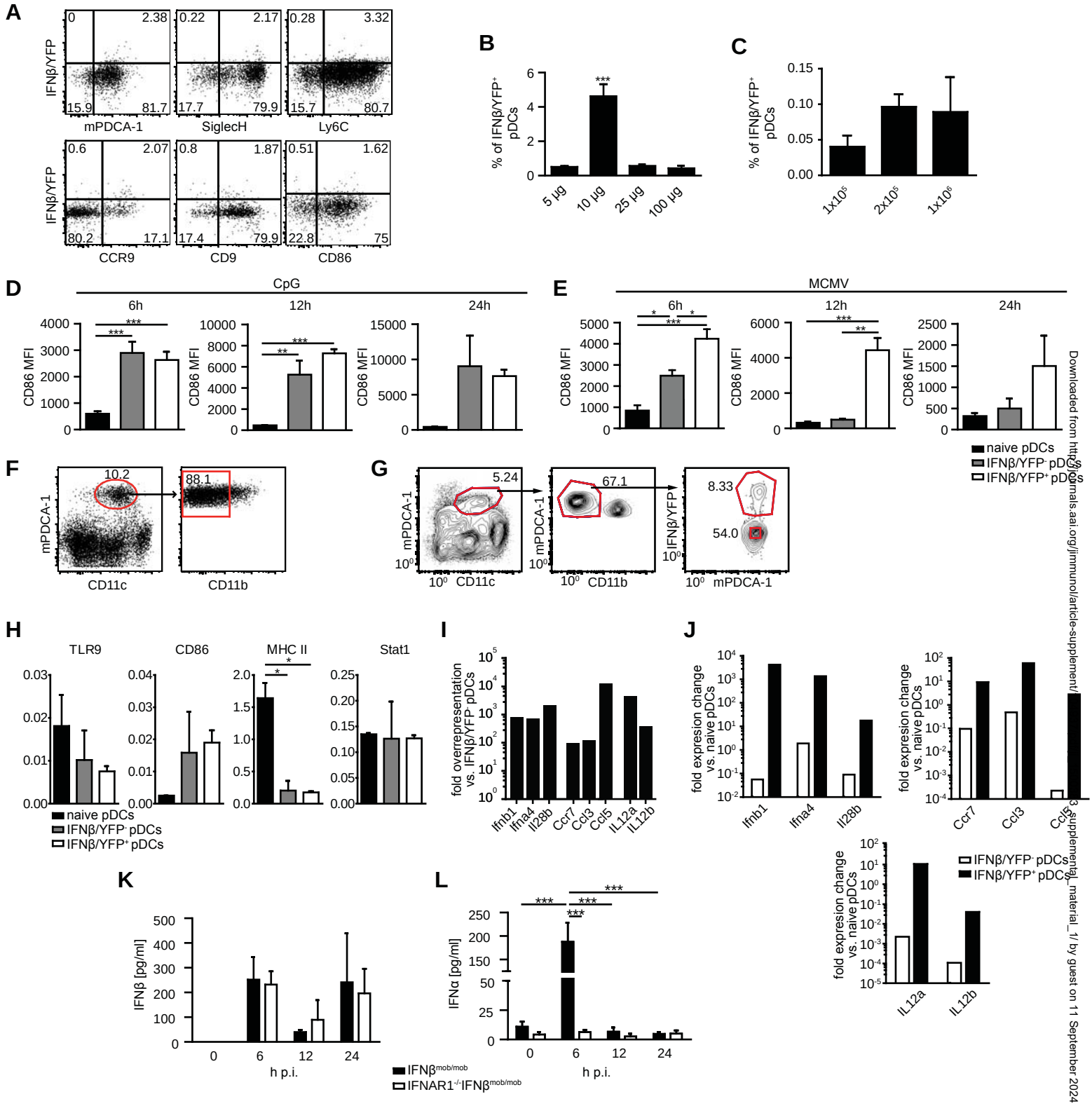

SUPPLEMENTAL FIGURE 1. IFNβ expression is restricted to a low frequent subset of splenic pDCs after MCMV infection or TLR9 stimulation.
(**A**) FACS analysis of IFNβ/YFP expression in BM-derived Flt3-L-cultured pDCs 6h after CpG stimulation. Cells were pre-gated on living CD3ε$^-$CD19$^-$CD11c$^+$CD11b$^-$B220$^+$ cells.
(**B**, **C**) IFNβ/YFP expression in splenic pDCs from IFNβ$^{mob/mob}$ mice 6h after injection of the indicated amount of CpG complexed to DOTAP (B) or infection with the indicated PFU dose of MCMV (C).
(**D**, **E**) MFIs for CD86 expression on naive pDCs from untreated mice (black bars), or IFNβ/YFP$^-$ pDCs (grey bars) and IFNβ/YFP$^+$ pDCs (white bars) from mice at indicated timepoints after injection of 10µg CpG complexed to DOTAP (D) or infection with 2x10^5 PFUs MCMV (E) were determined.
(**F**) Gating strategy for analysis of surface marker expression on splenic pDCs (as shown in Fig. 1E).
(**G**) Sort strategy for microarray analyses of splenic IFNβ/YFP$^+$ and IFNβ/YFP$^-$ pDCs after CpG injection (as shown in Fig. 3).
(**H**) Expression of genes associated with TLR9 activation in pDCs was analyzed by qRT-PCR. Expression is shown as $2^{-\Delta ct}$ values for naive pDCs from untreated mice (black bars), IFNβ/YFP$^-$ pDCs (grey bars) and IFNβ/YFP$^+$ pDCs (white bars).
(**I, J**) Differential expression of selected immune-relevant genes was verified by qRT-PCR, shown as fold overrepresentation of genes in IFNβ/YFP$^+$ vs. IFNβ/YFP$^-$ cells (I) or as fold expression change of IFNβ/YFP$^-$ (white bars) or IFNβ/YFP$^+$ cells (black bars) vs. naive pDCs (J). Mice were treated and pDCs were sorted as described in Fig. 3. All assays were performed in triplicate and gene expression was normalized to β-actin. One of three (H,I) or one of two independent experiments (J) are shown.
(**K, L**) Serum concentrations of IFNβ (K) and IFNα (L) in IFNβ$^{mob/mob}$ (black bars) and IFNAR1$^{-/-}$IFNβ$^{mob/mob}$ (white bars) mice at indicated timepoints after injection of 10 µg CpG complexed to DOTAP as determined by ELISA (PBL Assay Science, eBioscience, BioLegend). Shown is one representative of three independent experiments with ≥3 mice per time-point.
One-way ANOVA with Bonferroni post-test was used for statistical analyses. Bar graphs show mean +SEM. If not indicated otherwise, cells were pregated on living CD3ε$^-$CD19$^-$CD11b$^-$CD11c^{int}mPDCA-1$^+$ cells for FACS analysis and sorting.

Supplemental Table 1: Primer sequences

Gene	Forward	Reverse	Probe #
Ifna4	5'-tcaagccatccttgtgctaa-3'	5'-gtcttttgatgtgaagaggttcaa-3'	3
Ifnb1	5'-caggcaacctttaagcatcag-3'	5'-cctttgacctttcaaatgcag-3'	95
Actb	5'-tgacaggatgcagaaggaga-3'	5'-cgctcaggaggagvaatg-3'	106
Ccl5	5'-tgcagaggactctgagacagc-3'	5'-gagtggtgtccgagccata-3'	110
Ccl3	5'-tgcccttgctgttcttctct-3'	5'-gtggaatcttccggctgtag-3'	40
Ccr7	5'-ctccttgtcattttccaggtg-3'	5'-tggtattctcgccgatgtagt-3'	29
CD86	5'-gaagccgaatcagcctagc-3'	5'-cagcgttactatcccgctct-3'	107
MHCII	5'-gtggtgctgatggtgctg-3'	5'-ccatgaactggtacacgaaatg-3'	26
IL6	5'-gctaccaaactggatataatcagga-3'	5'-ccaggtagctatggtactccagaa-3'	6
Il28b	5'-tcagccctgaccaccatc-3'	5'-ctgtggcctgaagctgtgta-3'	33
Il12a	5'-ccatcagcagatcattctagacaa-3'	5'-cgccattatgattcagagactg-3'	49
Il12b	5'-tcttcaaaggcttcatctgcaa-3'	5'-acagcaccagcttcttcatca-3'	n.a.
Stat1	5'-aaatgtgaaggatcaagtcatgtg-3'	5'-catcttgtaattcttctagggtcttga-3'	15
TLR9	5'-gagaatcctccatctcccaac3'	5'-ccagagtctcagccagcac3'	79