Figure S1. Gating strategy for human pDC, mDC and naive CD4 T cell purification
We stained PBMC with CD3, CD14, CD16, CD19 PeCy7 (Lineage), HLA DR V450, CD4 ECD, CD11c FITC, CD45RA PE, CD27 APC antibodies. Among Lin.- cells, the further staining for CD4, CD11c and HLA DR discriminates between pDC and mDC. Among Lin. + cells CD4high we can discriminate naive T cells (CD45RA high CD27+). Cells are sorted by high speed cell sorting (Moflo, Coulter) (A). The analysis of sorted cells after sorting showed purity around 96% (B).

Figure S2. Plasmacytoid and myeloid dendritic cells from RR-MS patients and HD express similar level of TLR7/8 expression
Analysis of the expression of TLR-7 and TLR-8 in pDCs and mDCs freshly isolated from the blood of HD (n=8) and RR-MS patients (n=7) in RR stable phase by Affymetrix array. Error bars represent SD.

Figure S3. Th9 polarization mediated by cytokines is similarly induced in HD and RR-MS
Naive CD4 T cells derived from HD or RR-MS patients, were cultured for 5 days in the presence of anti-CD3 + anti-CD28 in absence (Th0) or presence of Th9 cocktail (TGF-β and IL-4). IL-9 production in culture supernatants, after 24 hours of restimulation, was measured by ELISA. Error bars represent SD. Paired t-test was used to compare no cytokines and Th9 cocktail. Data are represented as mean ± SD of 5 donors. Unpaired t-test was used to compare HD and RR-MS. ** P < 0.005, *** P < 0.001.

Figure S4. IL-4 does not influence disease severity in RR-MS
Clinical parameters ARR (A) and PI (B) and the levels of NfL in CSF (C) were analysed in RR-MS subjects with undetectable (und) or detectable (det) IL-4. Correlation between levels of IL-9 and IL-4 in CSF was performed using a Pearson correlation.

Figure S5. Human Th subsets express IL-9 receptor
RT-PCR for expression of IL9 receptor (IL9R) mRNA in naive T cells differentiated with anti-CD3 + anti-CD28 in Th0, Th1, Th2, Th9, Th17 and Treg condition for 5 days. RT-PCR was performed after 24h of re-stimulation with anti-CD3 + anti-CD28 and Ct values were normalized to mRNA of ribosomal protein L-34. Th0, absence of any polarizing cytokine; Th1, addition of IL-12; Th2, addition of IL-4; Th17, addition of IL-1α, IL-6, TGF-β and IL-23; Treg, TGF-β and IL-2. Data are represented as mean ± SD of 5 donors.

Figure S6. IL-9 specifically modulates IL-17 production by Th17 cells
Naive CD4 T cells were stimulated with anti-CD3 + anti-CD28 in presence of Th17 polarizing cytokines for 5 days. At day 4 cells were treated or not with IL-9 for the last period of culture. IL-17 protein in the supernatants after 24 hours of re-stimulation with antiCD3-28 was analysed by ELISA for IL-17, TNF-α, IL-6, IL-10 and IFN-γ; Paired t-test was used to compare sample conditions. * P < 0.05.
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