Supplementary Material

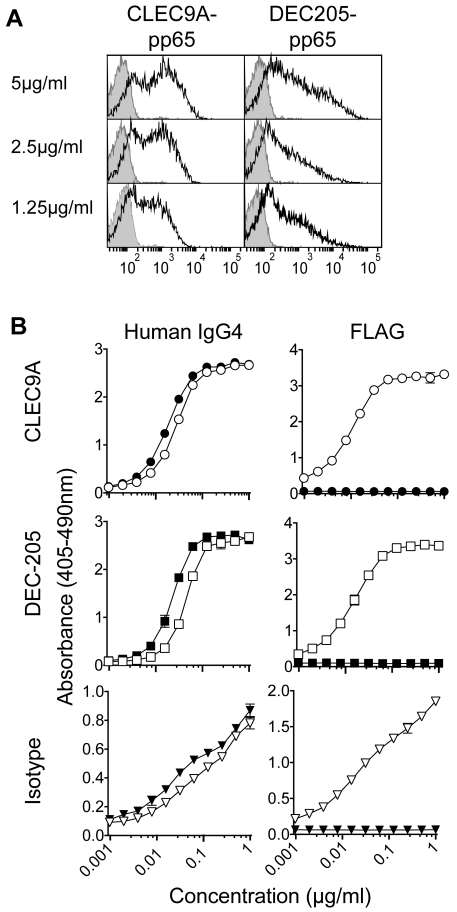

Figure S1. *Validation of purified human chimaeric Ab-pp65 fusion proteins*. (**A**) Ability of purified Ab-pp65 fusion proteins to retain binding to target receptor was assessed by staining of 293F cells transfected with recombinant full-length CLEC9A with anti-CLEC9A-pp65 (black, left panel) or full-length recombinant DEC-205 with anti-DEC-205-pp65 (black, right panel) over a range of concentrations compared to the isotype control (grey). Ab-pp65 fusion proteins were detected with anti-human IgG4-biotin and streptavidin-PE and analyzed by flow cytometry (**B**) Validation of purified human chimeric Ab (open symbols) and Ab-pp65 fusion proteins (closed symbols) by ELISA. Ab or Ab-pp65 fusion proteins were incubated on plates coated with CLEC9A peptide, DEC-205 recombinant protein or β-gal and were detected using anti-human IgG4-biotin and streptavidin-HRP (left panel) or anti-FLAG-HRP Ab (right panel).

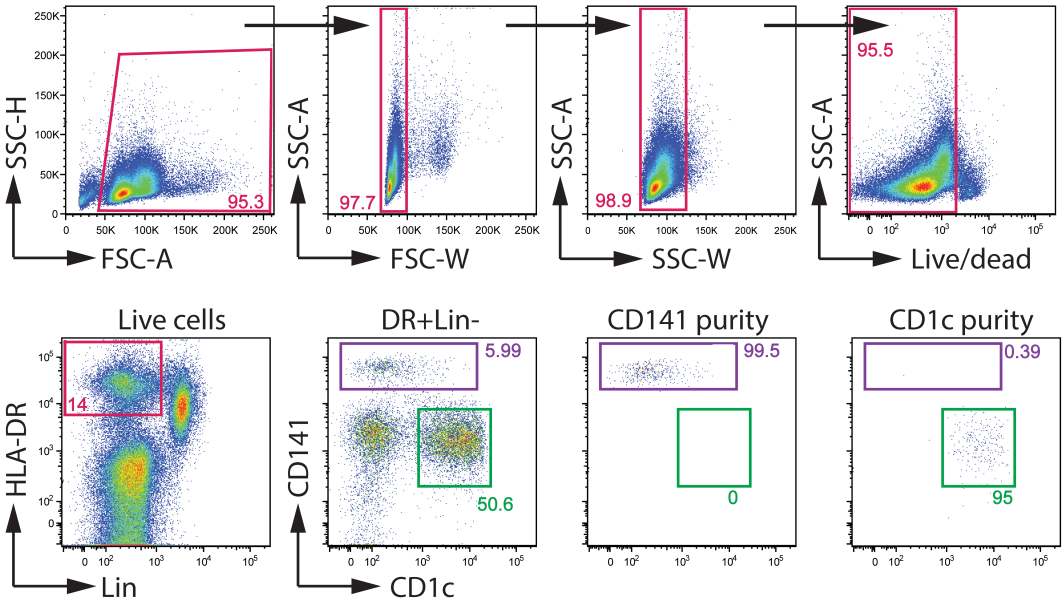

Figure S2. *Gating strategy for sorting of human blood DC.* PBMC were isolated using a Ficoll Hypaque density gradient and cDC were enriched using the Myeloid DC enrichment kit (Stem Cell). The enriched cells were stained with live/dead aqua, HLA-DR, CD1c, CD141 and lineage markers (lin); CD3, CD14, CD16, CD19, CD20, CD56. Cells were sorted using a live cell gate and HLA-DR^{+}lin^{-} and according to expression of CD141 and CD1c.

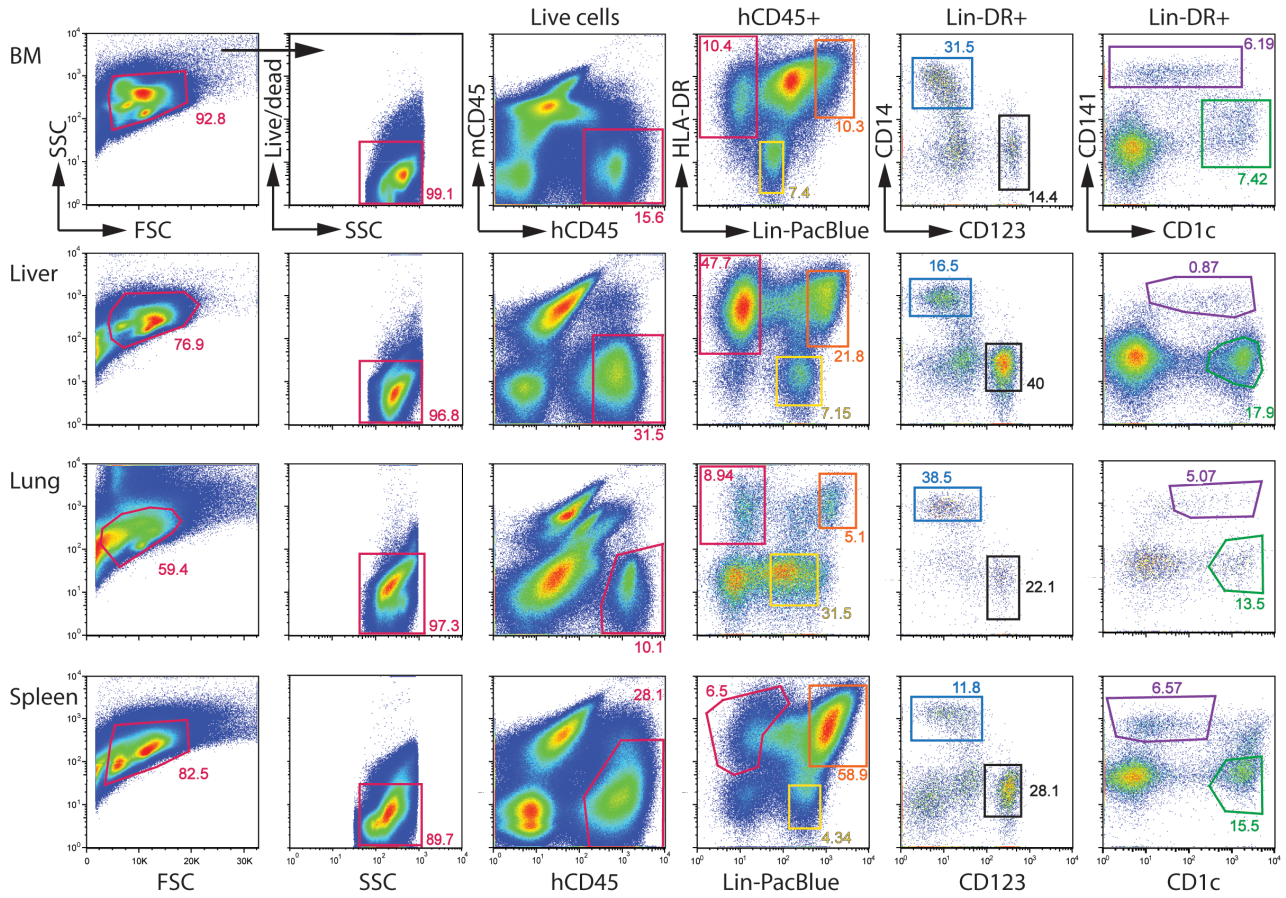

Figure S3. *Gating strategy for identification of human DC in engrafted NSG-A2 humanized mice.* BM, liver, lung and spleen tissue were harvested 2h post-injection with 5µg of Alexa Fluor (AF)488-conjugated human chimaeric Ab and leukocytes were stained with live/dead, HLA-DR, CD141, CD1c, CD123, CD14 and lineage markers; mouse (m)CD45, human (h)CD45, CD3, CD16, CD19 and CD20. In addition, CD14 was included in the lineage markers for identification of $CD141^+$ DC and $CD1c^+$ DC subsets. Human cells were identified as live $hCD45^+$ $mCD45^-$ and gated according to expression of lineage markers (lin); T cells (yellow):$HLA\text{-}DR^-lin^+$, B cells (orange):$HLA\text{-}DR^+lin^+$, Monocytes (blue): $HLA\text{-}DR^+lin^-CD14^+$, pDC (black): $HLA\text{-}DR^+lin^-CD123^+$, CD141 DC (purple): $HLA\text{-}DR^+lin^-CD14^-CD141^+$, CD1c DC (green): $HLA\text{-}DR^+lin^-CD14^-CD1c^+$.

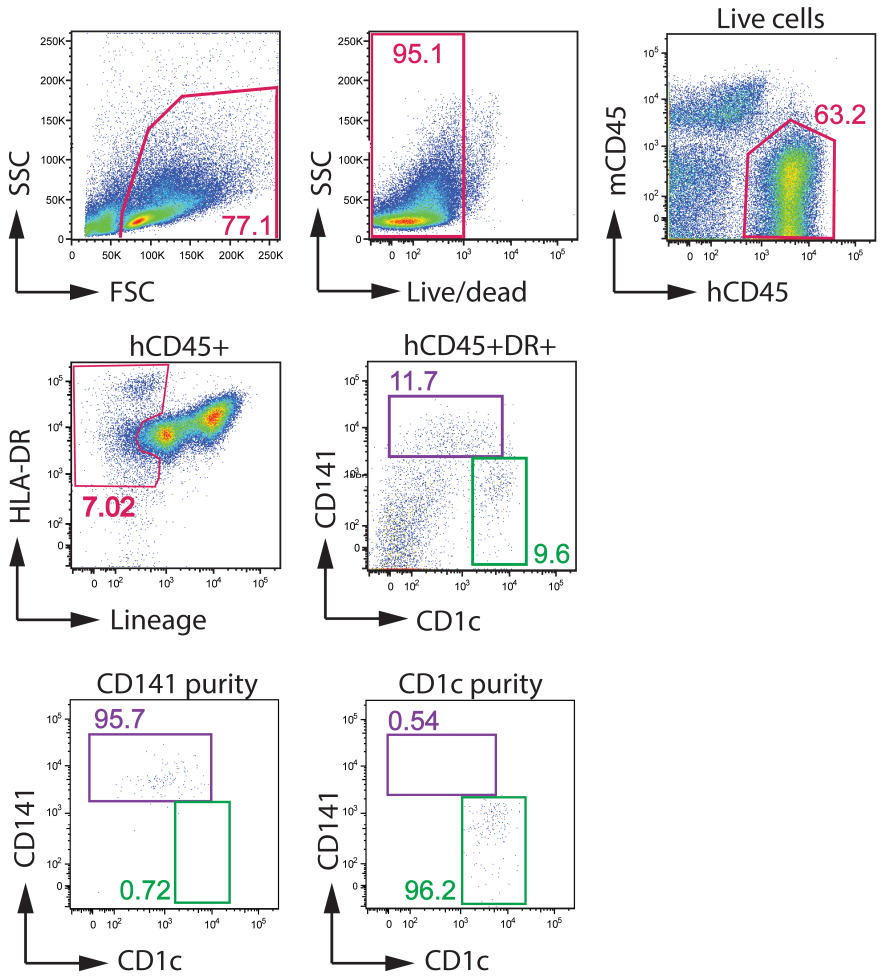

Figure S4. *Gating strategy for sorting of human DC from spleen of engrafted huNSG-A2 mice.* Spleen tissue from engrafted and Flt3L-injected NSG-A2 humanized mice was harvested 24h post-injection with 10µg Ab-pp65 and 50µg polyI:C. Leukocytes were isolated using density gradient centrifugation and stained with live/dead aqua, human (h)CD45, mouse (m)CD45, HLA-DR, CD123, CD1c, CD141 and lineage markers (lin); CD3, CD14, CD16, CD19, CD20. Human DC were identified using live cell gates and were $hCD45^{+}mCD45^{-}HLA\text{-}DR^{+}lin^{-}$ and sorted according to expression of CD1c and CD141.