

## Supplemental Figure 8: Characterization of anti-human CCR8 antibody

(A) Binding of anti-human CCR8 antibodies CCR8.1-IgG1-nf ( $EC_{50}$ = 0.35 nM), CCR8.2-IgG1-nf ( $EC_{50}$ =8.41 nM), or isotype control to *in vitro* activated human T<sub>regs</sub>. (B) CCL1 calcium flux assay in the presence of CCR8.1-IgG1-nf ( $IC_{50}$ = 0.27 nM), CCR8.2-IgG1-nf ( $IC_{50}$ =1.33 nM), or isotype control. (C) CD16 crosslinking luciferase reporter assay with activated human T<sub>regs</sub> in the presence of CCR8.1-IgG1-nf ( $EC_{50}$ = 2.48 pM), CCR8.2-IgG1-nf ( $EC_{50}$ =31.0 pM), or IgG1-nf isotype control. (D) CD16 crosslinking luciferase reporter assay on human CCR8 expressing Raji cells in the presence of CCR8.1-IgG1-nf ( $EC_{50}$ =0.26 pM), CCR8.1-IgG1 ( $EC_{50}$ =2.37 pM), or IgG1-nf isotype control. (E) Comparison of CD16 crosslinking between CCR8.1-IgG1-nf ( $EC_{50}$ = 1.08 pM) and CCR8.1-IgG1-inert, with isotype control. Non-linear regression (log[agonist] vs. response) was applied (A-E).