

Supplementary Figures for *Scientific Reports*:

STING agonists activate latently infected cells and enhance SIV-specific responses *ex vivo* in naturally SIV controlled cynomolgus macaques

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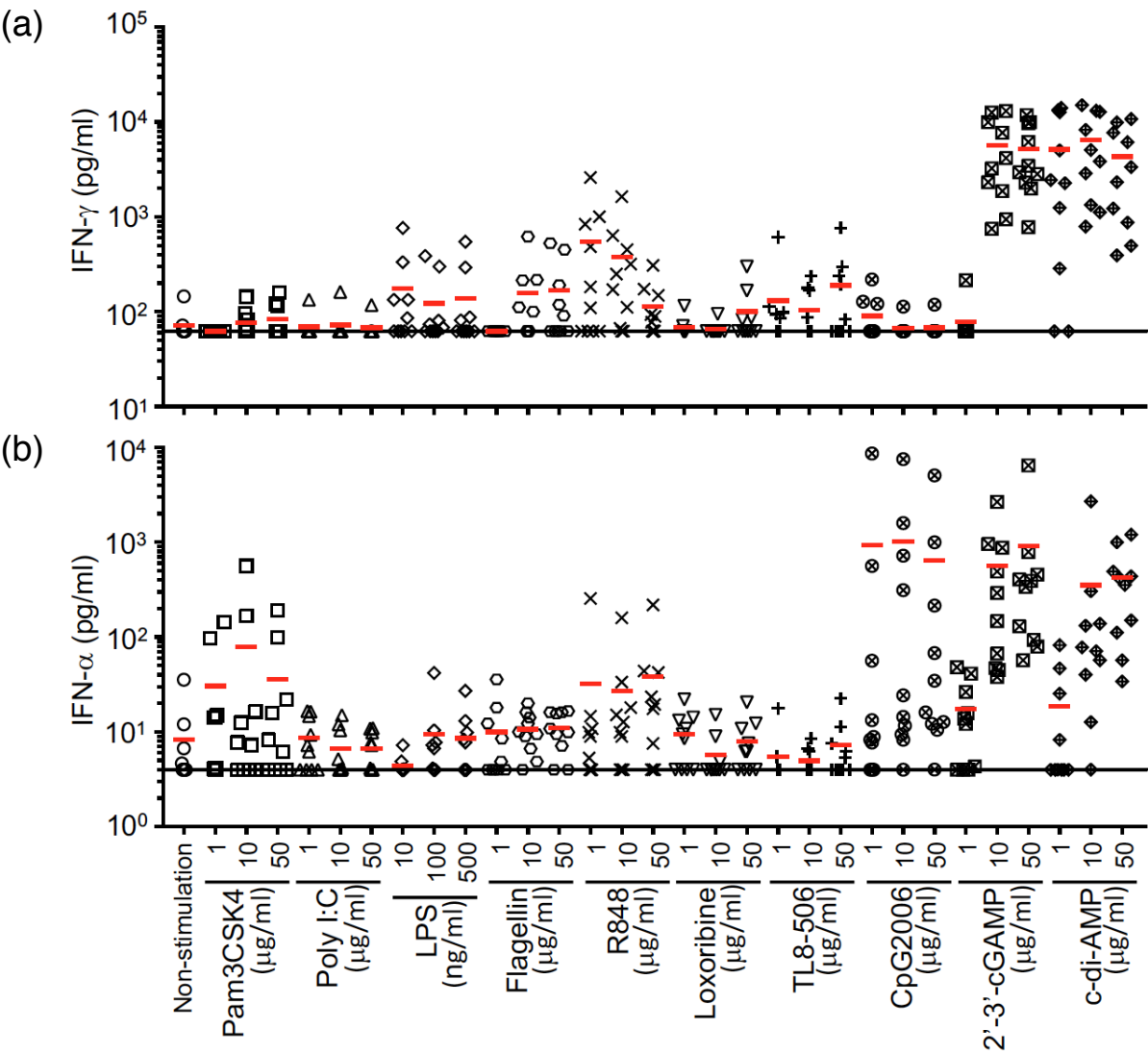
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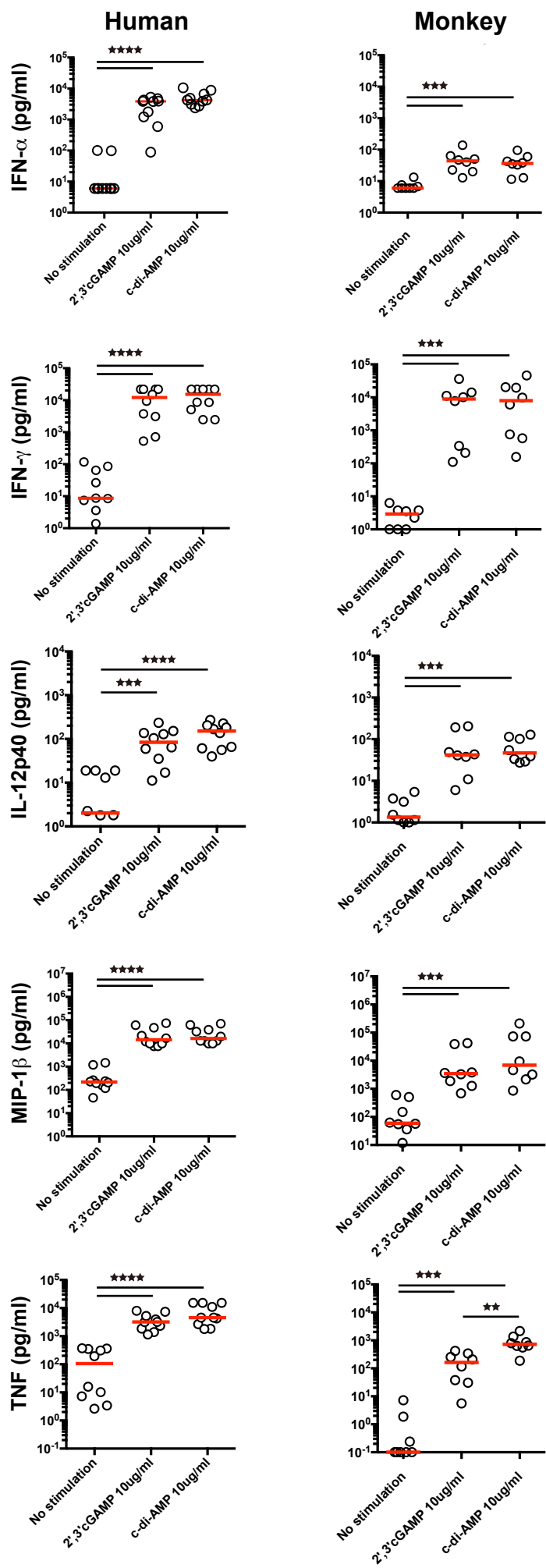
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Supplementary Figure S1



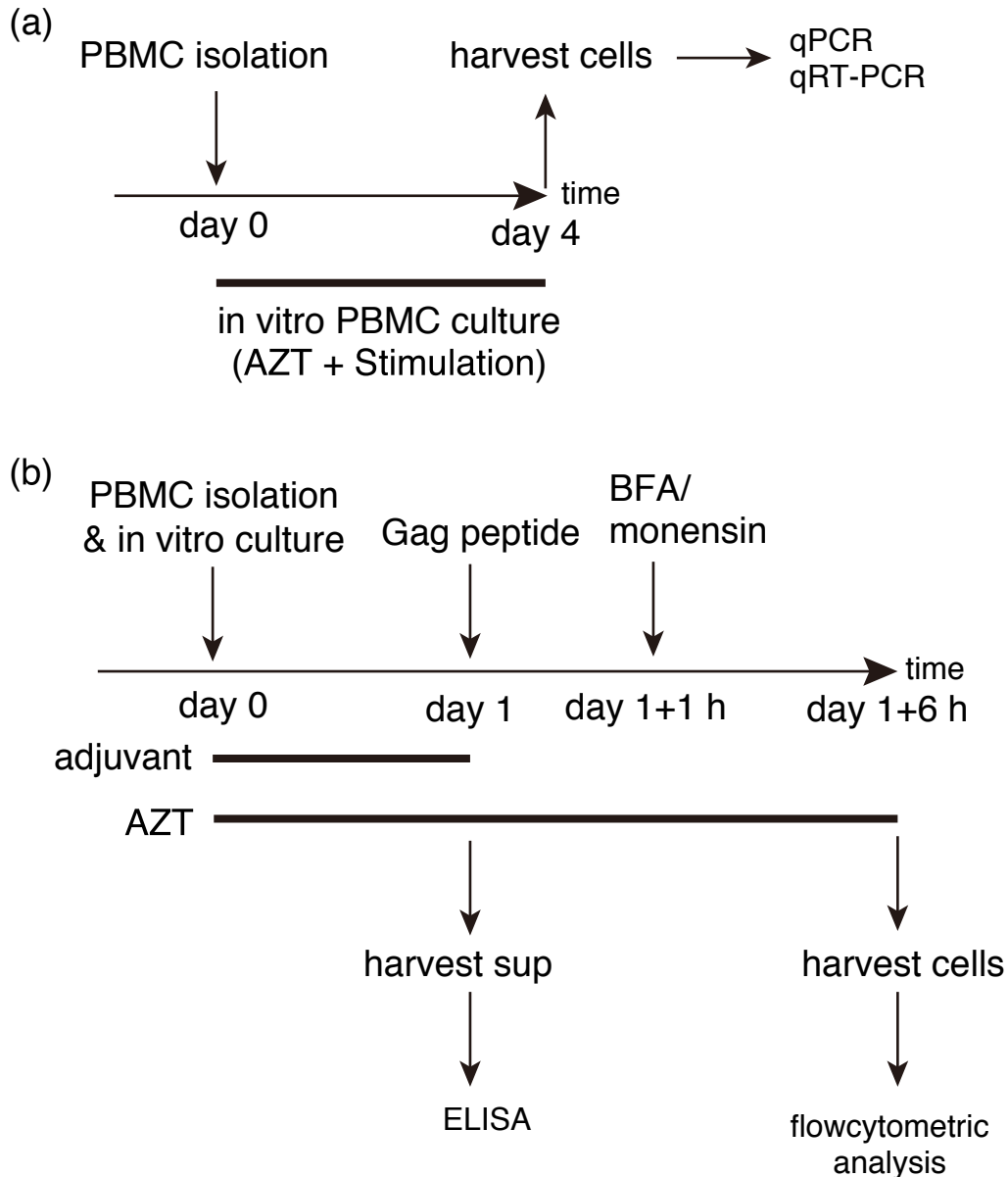
Optimization of adjuvant concentrations.
PBMCs isolated from cynomolgus monkeys were treated with indicated adjuvant for 24 hrs, then cytokines in culture supernatant were analyzed by ELISA. a) levels of IFN- γ were plotted. b) levels of IFN- α were plotted. Bars indicate median values.

Supplementary Figure S2



Comparison of the effect of STING ligands for human or monkey PBMCs. PBMCs isolated from healthy human or cynomolgus monkeys were treated with indicated STING ligands for 24 hrs, then cytokines in culture supernatant were analyzed by ELISA. Bars indicate median values.

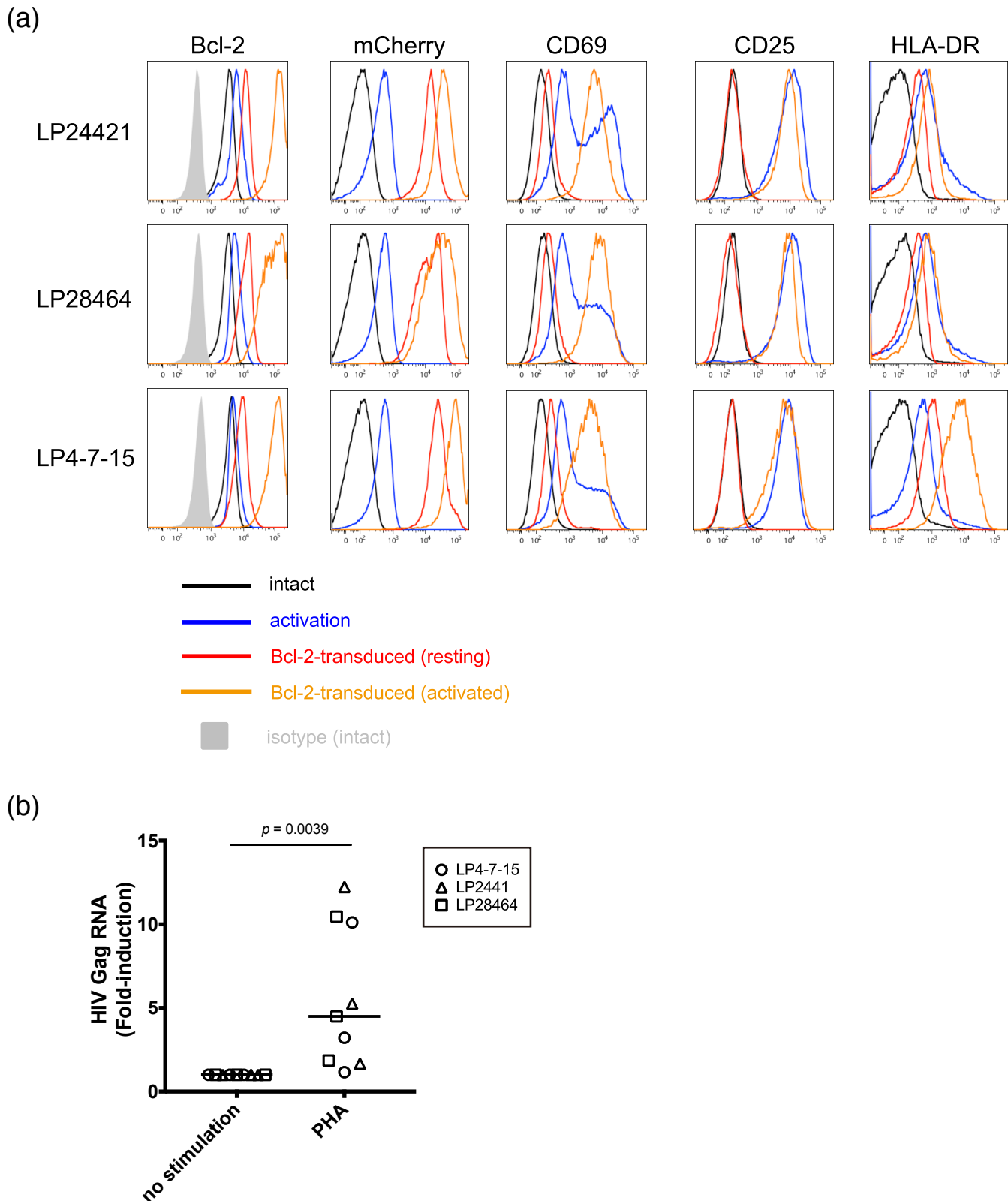
Supplementary Figure S3



Schematic representation of experimental design.

(a) Evaluation of SIV Gag DNA or RNA in infected cells or culture supernatants. Isolated PBMCs were cultured with AZT and R848 or STING ligand for 4 days. After that, the cells were harvested, and the culture supernatants were collected, and then the cellular DNA or RNA and supernatant RNA were purified. These samples were evaluated by qPCR or qRT-PCR. (b) Identification of the function of adjuvant-stimulated PBMCs. Isolated PBMCs were cultured with AZT and adjuvants for 24 hours. After that, some cell culture supernatant was collected for the analysis of cytokine/chemokine expression by ELISA. These cells were stimulated with SIV Gag peptides for 24 hours and BFA and monensin were added after 1 hour. After SIV Gag peptide stimulation, the functions of these cells were evaluated by flow cytometry.

Supplementary Figure S4



Establishment of HIV genes containing Bcl-2-transduced human CD4⁺ T cells

a) Resting and activated status of parental and Bcl-2-expressing human CD4⁺ T cells. CD4⁺ cells isolated from PBMCs from patients were stimulated with CD3/CD28, and then cultured for 3 days. Then, cells were infected with Bcl-2/mCherry-expressing lentivirus and cultured for 3 days. mCherry-positive cells were sorted and expanded repeatedly for 10 weeks. Expression of Bcl-2/mCherry and activation markers in established Bcl-2-expressing CD4⁺ cells were analysed by FACS.

b) Established CD4⁺ T cells were stimulated with PHA for 4 days, then the levels of cellular HIV Gag RNA were analyzed by qPCR.

