

Supplementary Materials for
Therapy with CTLA4-Ig and an antiviral monoclonal antibody controls
chikungunya virus arthritis

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The PDF file includes:

- Fig. S1. RANKL and OPG expression in CHIKV-infected joints.
- Fig. S2. Foot swelling with CTLA4-Ig therapy administered before or during CHIKV infection.
- Fig. S3. Flow cytometry analysis of APC activation in the spleen and ankles of mice on day 7 after infection with CHIKV.

Other Supplementary Material for this manuscript includes the following:
(available at www.sciencetranslationalmedicine.org/cgi/content/full/9/375/eaah3438/DC1)

Table S1 (Microsoft Excel format). Primary data.

SUPPLEMENTARY FIGURES

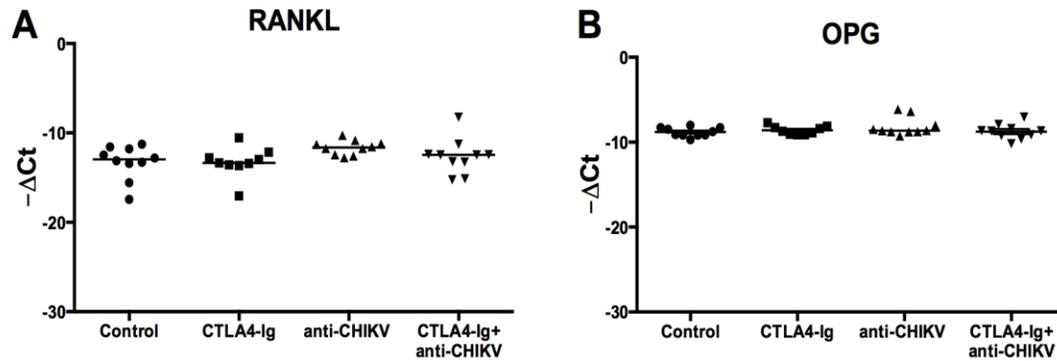


Fig. S1. RANKL and OPG expression in CHIKV-infected joints. Mice were inoculated with 10^3 FFU of CHIKV via a subcutaneous route. At day 3 after infection, a single IP injection of 600 μg of isotype control antibody, 300 μg of CTLA4-Ig, 300 μg of anti-CHIKV mAb, or a combination of 300 μg CTLA4-Ig and 300 μg anti-CHIKV mAb was administered. On day 7 after infection, RNA was isolated from the left foot. RANKL (**A**) and OPG (**B**) expression were measured by qRT-PCR and normalized to GAPDH. Data were pooled from two independent experiments ($n = 10$ animals per group) and analyzed by ANOVA with a multiple comparisons test. $P > 0.1$ for all groups.

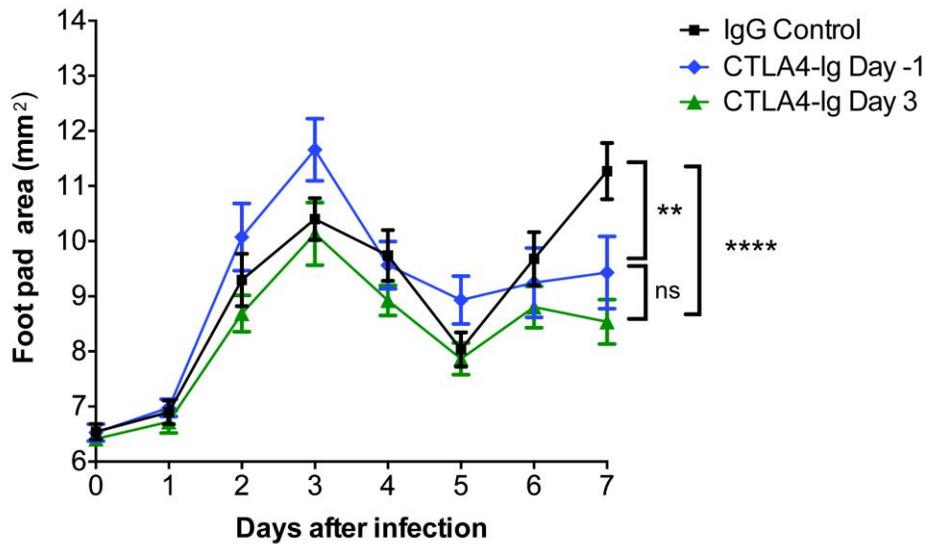


Fig. S2. Foot swelling with CTLA4-Ig therapy administered before or during CHIKV infection. Five week-old mice were inoculated with 10^3 FFU of CHIKV via a subcutaneous route. (A) Foot swelling (area in mm^2) from day 0 through day 7 in mice receiving a single IP injection of $300 \mu\text{g}$ of isotype control antibody or $300 \mu\text{g}$ of CTLA4-Ig at day -1 or day 3. Data are pooled from two independent experiments ($n = 9$ or 10 animals per group). Data represent the mean \pm SEM. **, $P < 0.005$; **** $P < 0.0001$ (2-way ANOVA with multiple comparisons test).

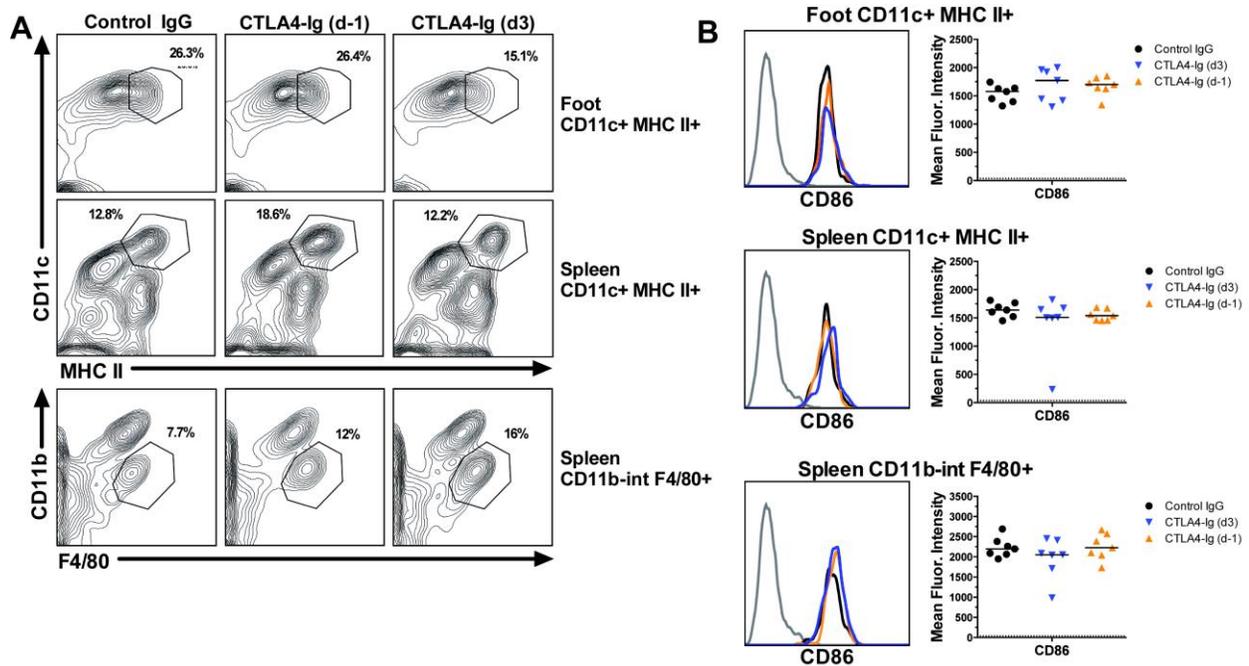


Fig. S3. Flow cytometry analysis of APC activation in the spleen and ankles of mice on day 7 after infection with CHIKV. Mice were inoculated with 10^3 FFU of CHIKV via a subcutaneous route. Mice received at day 3 a single IP injection of 300 μ g of isotype control antibody 300 μ g of CTLA4-Ig on either day -1 or day 3 after infection. (A) Representative contour plots showing the percentages of CD11c⁺MHCII⁺ dendritic cells and CD11b^{intermediate}F4/80⁺ macrophages in the left foot and spleen of infected animals from each treatment group. (B) Representative histograms (left panel) and geometric mean fluorescence intensity of CD86 expression on CD11c⁺MHCII⁺ dendritic cells and CD11b^{intermediate}F4/80⁺ macrophages in the left foot and spleen of infected animals from each treatment group. Results are pooled from two independent experiments with 3 to 4 mice per group per experiment. No statistically significant differences were observed in the treatment groups (Kruskal-Wallis with Dunn's post-hoc analysis).