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Supplementary Materials for

CD8$^+$ T cells mediate protection against Zika virus induced by an NS3-based vaccine

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This PDF file includes:

Figure S1 related to figure 2

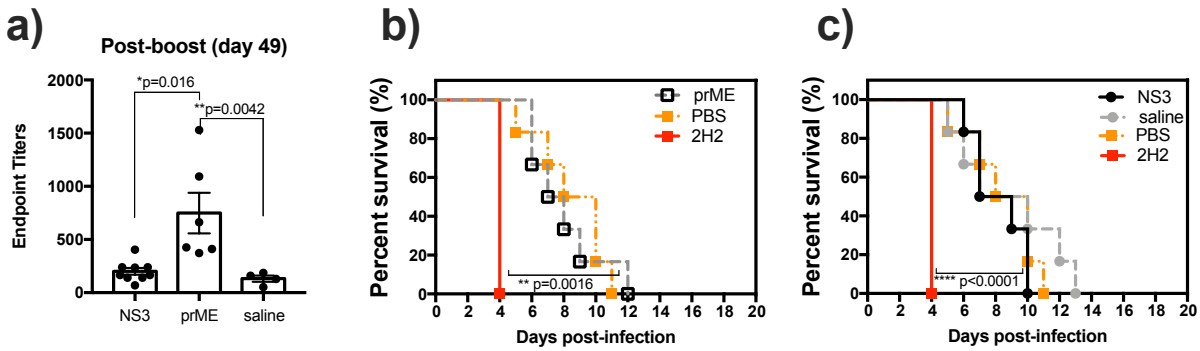

Figure S1 related to Figure 2. Endpoint titers of sera from NS3- or prM-E-vaccinated wild-type C57BL/6 mice and survival of DENV2-infected AG129 mice after transfer of serum from wild-type C57BL/6 mice vaccinated with ZIKV NS3 or prM-E

Groups of wild-type C57BL/6 mice were injected i.m. with 10 µg of NS3 (n=10) or prM-E (n=6) vaccines or with saline (n=4) and boosted in the same manner on day 28. (**A**) On day 49, sera from prM-E-vaccinated mice were collected to quantify anti-ZIKV IgG titers using a ZIKV E protein ELISA. The endpoint titers were calculated and sera from NS3-vaccinated or saline-injected mice were used as controls. Data are presented as the mean ± SEM, with individual mice shown as circles. (**B and C**) Sera were pooled and injected i.p. into groups of 5–6-week-old naïve AG129 mice (n=6). As positive and negative controls, additional groups of naïve AG129 mice (n=6) were injected i.p. with 15 µg of anti-DENV prM-E Ab 2H2 or PBS, respectively. One day later, all mice were infected i.v. with 10^5 FFU DENV2 221 and survival was monitored. Data are pooled from two independent experiments. Survival data were compared using the Gehan–Breslow–Wilcoxon test (2H2 *vs* NS3 or prM-E groups). The nonparametric Kruskal–Wallis test was used to compare three groups and the Mann–Whitney test was used to compare two groups

Figure S2 related to figure 3

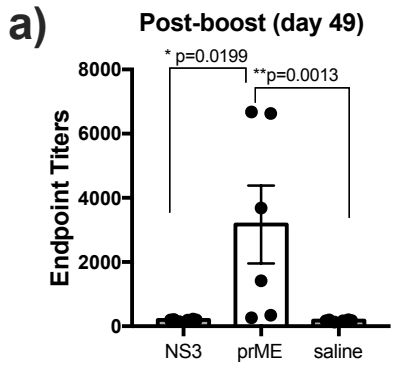

Figure S2 related to Figure 3. Endpoint titers of sera from NS3- or prM-E-vaccinated HLA-B*0702 *Ifnar1*$^{-/-}$ mice

(**A**) Groups of HLA-B*0702 *Ifnar1*$^{-/-}$ mice were vaccinated i.m. with 10 μg of NS3 (n=6) or prM-E (n=6) vaccines or with saline (n=5) and boosted in the same manner on day 28. On day 49, sera were collected to quantify anti-ZIKV IgG using ELISA against ZIKV E protein. The endpoint titers were calculated. Data are presented as the mean ± SEM, with individual mice shown as circles. The nonparametric Kruskal–Wallis test was used to compare three groups and the Mann–Whitney test was used to compare two groups.

Figure S3 related to figure 4

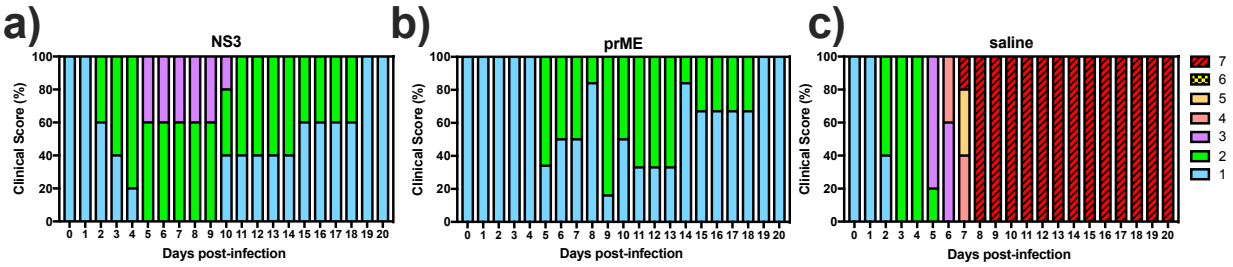

Figure S3 related to Figure 4. Clinical scores of NS3- or prM-E-vaccinated HLA-B*0702 *Ifnar1*$^{-/-}$ mice after lethal infection with ZIKV

(**A–C)** Groups of HLA-B*0702 *Ifnar1*$^{-/-}$ mice were vaccinated i.m. with 10 µg of NS3 (n=6) or prM-E (n=6) vaccines or with saline (n=5) and boosted in the same manner on day 28. On day 49, mice were lethally infected r.o. with 10^4 FFU of ZIKV SD001. Clinical signs (appearance, mobility) were monitored daily, and disease was scored on a 7-point scale: 1, healthy; 2, slightly ruffled coat around head and neck; 3, ruffled coat over the entire body; 4, severely ruffled coat and slightly closed eyes; 5, sick with closed eyes and slow movement (mice were euthanized); 6, no movement and slow breathing; 7 dead.

Figure S4 related to figure 6

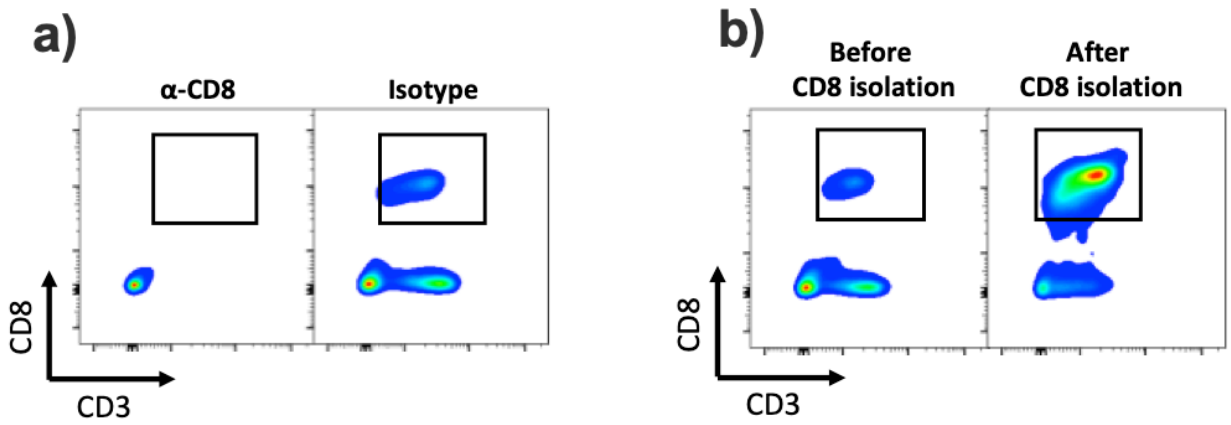

Figure S4 related to Figure 6. Characterization of CD8-depletion and CD8+ T cell isolation of NS3-vaccinated or saline-injected HLA-B*0702 *Ifnar1*$^{-/-}$ mice.

HLA-B*0702 *Ifnar1*$^{-/-}$ mice were vaccinated with 10 µg of NS3 vaccine (n=11) or saline (n=12), boosted in the same manner on day 28, and then treated i.p. with CD8+ cell-depleting Ab (2.43) or isotype control Ab prior to ZIKV SD001 infection on day 49. **(A)** A representative dot-plot of CD8+ T cell depletion in the spleen was represented on day 52 by flow cytometry. HLA-B*0702 *Ifnar1*$^{-/-}$ mice were vaccinated as described for (A). On day 49, CD8+ T cells were isolated from pooled spleens of vaccinated-mice and **(B)** a representative dot-plot of the purity of transferred CD8+ T cells was presented.