SUPPORTING INFORMATION

Supporting Information Figure 1. Graphs represent donor Treg cell recovery (as % of total Treg cells) from secondary lymphoid organs of individual recipient mice at (A) 1 week, (B) 4 weeks, or (C) 12 weeks following transfer. Symbols represent individual recipients with lines representing means. These data are the complete set from which representative flow plots are shown in Fig. 1A–C. Data are pooled from 2 (A), 4 (B), or 3 (C) independent experiments. CervLN, cervical LN; gutLN, gut-draining LN; skinLN, skin-draining LN.

Supporting Information Figure 2. Non-cervLN Treg cells do not accumulate within cervLNs following adoptive transfer. FACS-purified skinLN and gutLN Treg cells from Foxp3-GFP reporter mice were transferred into congenic Foxp3-GFP recipient mice. At 1, 4, and 12 weeks following transfer secondary lymphoid organs were harvested and analyzed by flow cytometry for the presence of congenically-marked donor Treg cells. Enrichment factors were calculated as in Fig. 1E. Graphs represent mean ± SEM of enrichment factors for indicated recovery sites and time points for skinLN (A) and gutLN (B) donors. Statistical analyses by two-way repeated measures ANOVA demonstrate significant difference between recovery sites ($p<0.0001$) for skinLN donor Treg cells with specific p values from Bonferroni post-test as shown (** $p<0.01$, *** $p<0.001$ compared with recovery from skinLN at same time point) but no significance for gutLN donor Treg cells. Data are pooled from 3 recipients for each time point for skinLN donor or 3 recipients for 1 and 4 week and 6 recipients for 12 week time points for gutLN donor in two

independent experiments. CervLN, cervical LN; gutLN, gut-draining LN; skinLN, skin-draining LN.

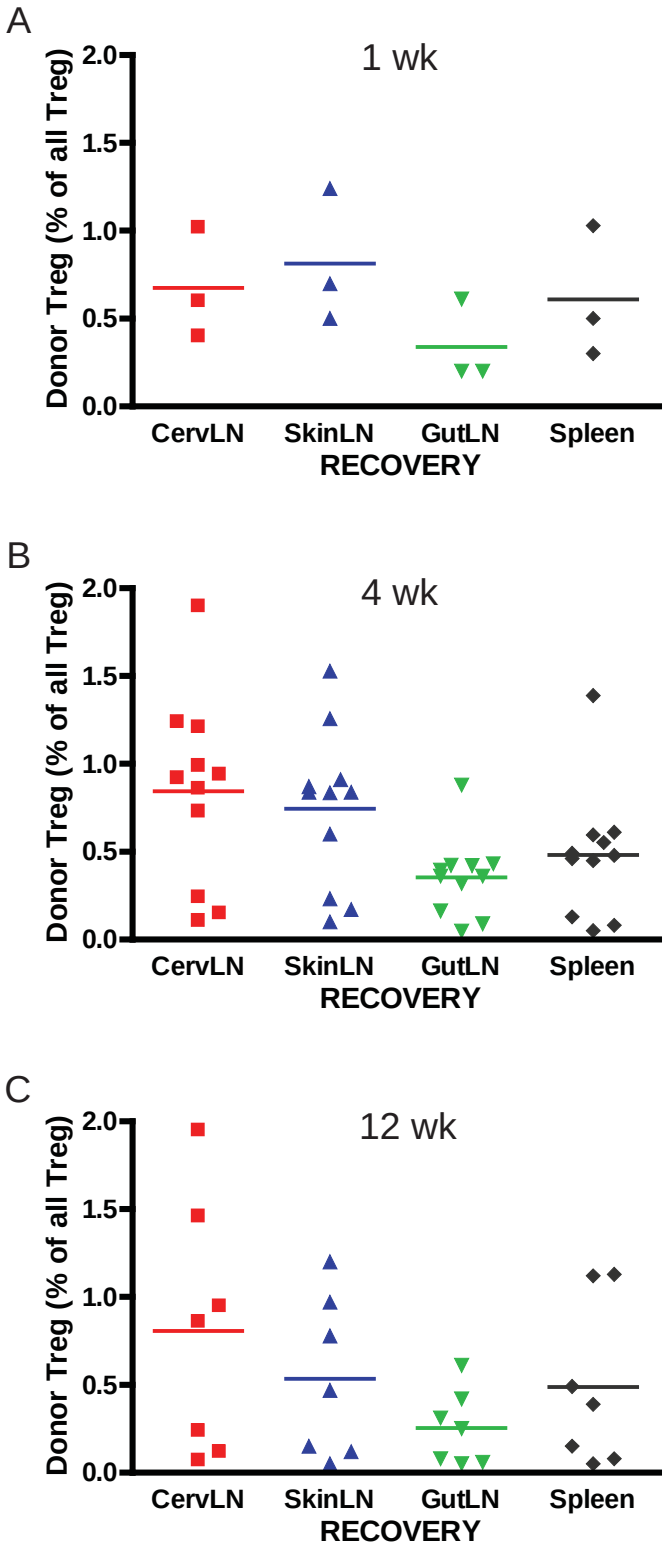
Supporting Information Figure 3. Tconv cells do not accumulate site-specifically following adoptive transfer. FACS-purified gutLN and non-gutLN (pooled cervLN and skinLN) Tconv cells from Foxp3-GFP reporter mice were transferred into congenic Foxp3-GFP recipient mice. At 4 and 12 weeks following transfer secondary lymphoid organs were harvested and analyzed by flow cytometry for the presence of congenically-marked donor Tconv cells. Enrichment factors were calculated as in Fig. 1E. Graphs represent mean ± SEM of enrichment factors for indicated recovery sites and time points for gutLN (A) and non-gutLN (B) donors. Statistical analyses by two-way repeated measures ANOVA demonstrate no significant differences for either donor Tconv cell group. Data are pooled from 3 recipients of gutLN Tconv cells at each time point or 3 (4 week) or 4 (12 week) recipients of non-gutLN Tconv cells in two independent experiments. CervLN, cervical LN; gutLN, gut-draining LN; skinLN, skin-draining LN.

Supporting Information Figure 4. Graphs represent $CD69^+$ (A) and $CD69^-$ (B) donor Treg cell recovery (as % of total Treg cells) from secondary lymphoid organs of individual recipient mice at 4 weeks post-transfer. Symbols represent individual recipients with lines representing means. These data are the complete set from which representative flow plots are shown in Fig. 2C. CervLN, cervical LN; gutLN, gut-draining LN; skinLN, skin-draining LN.

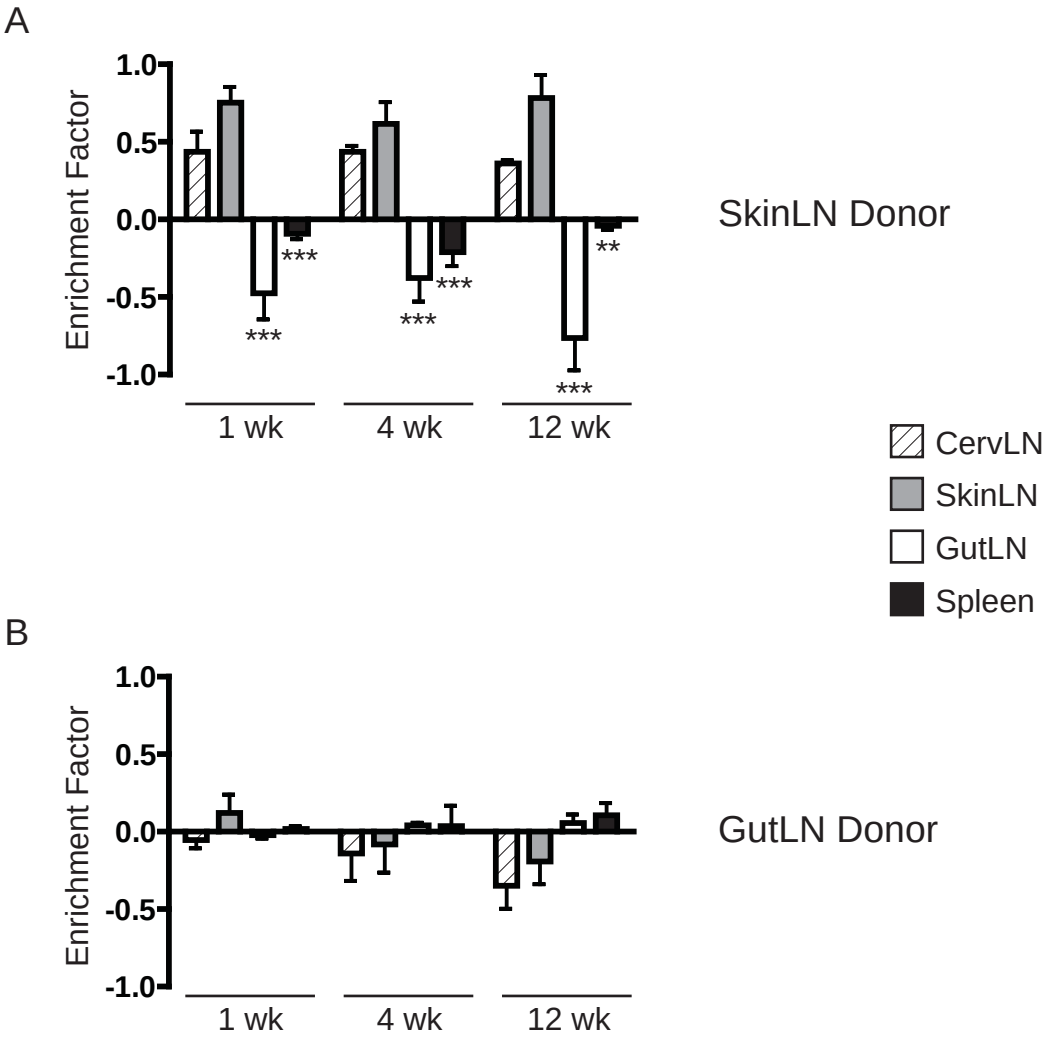
Supporting Information Figure 5. No enrichment advantage of $CD69^+$ cervLN Treg cells at non-cervLN recovery sites. Graph represents mean ± SEM of non-site-specific enrichment

factors (i.e., enrichment factor for cervLN donor Treg cells recovered at non-cervLNs) for $CD69^+$ and $CD69^-$ donor Treg cells from 7 recipients in 2 independent experiments or from bulk cervLN donor Treg cells from 4 week time point in Fig. 1E. CervLN, cervical LN; gutLN, gut-draining LN; skinLN, skin-draining LN.

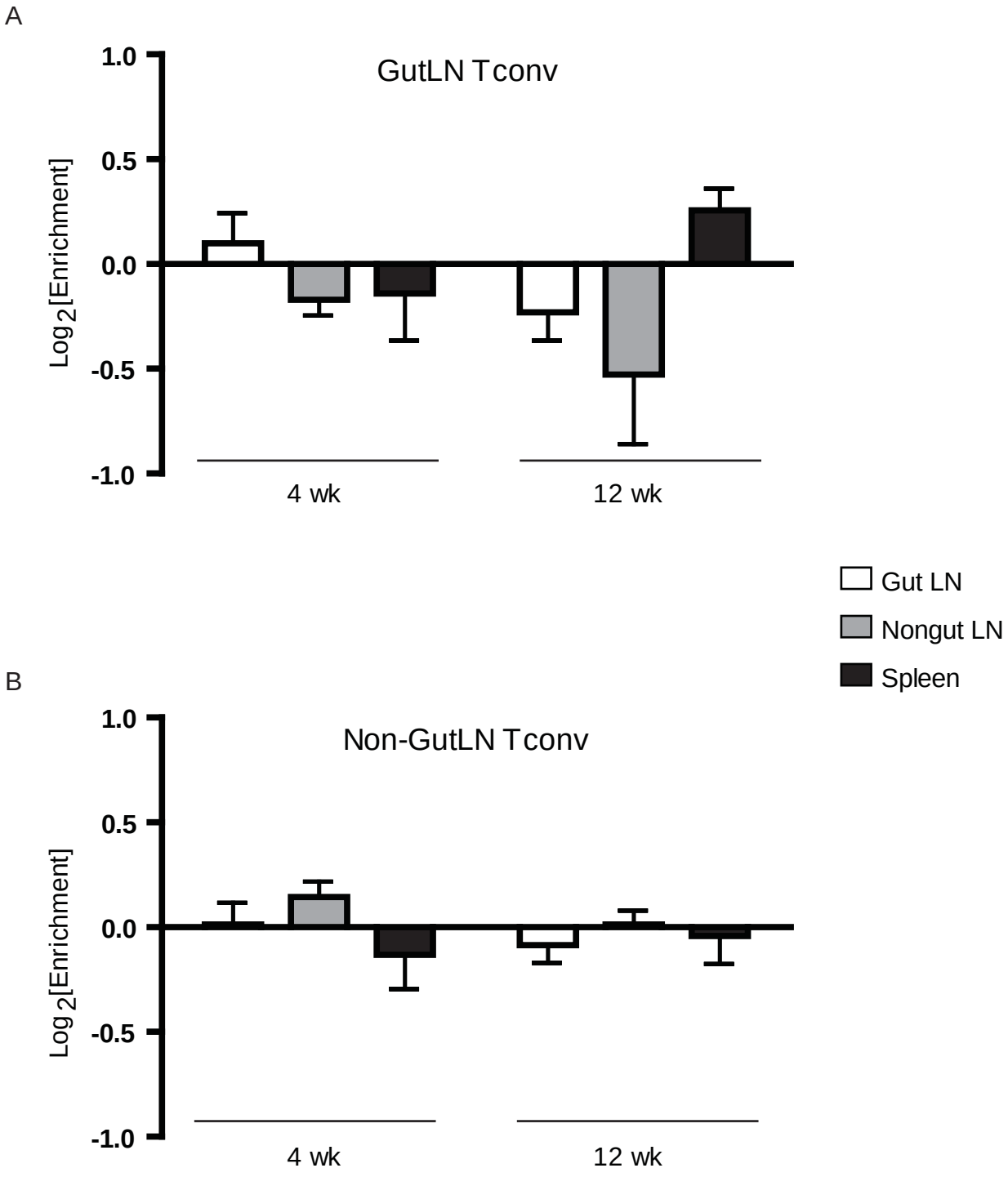
SUPPORTING INFORMATION FIGURE 1.



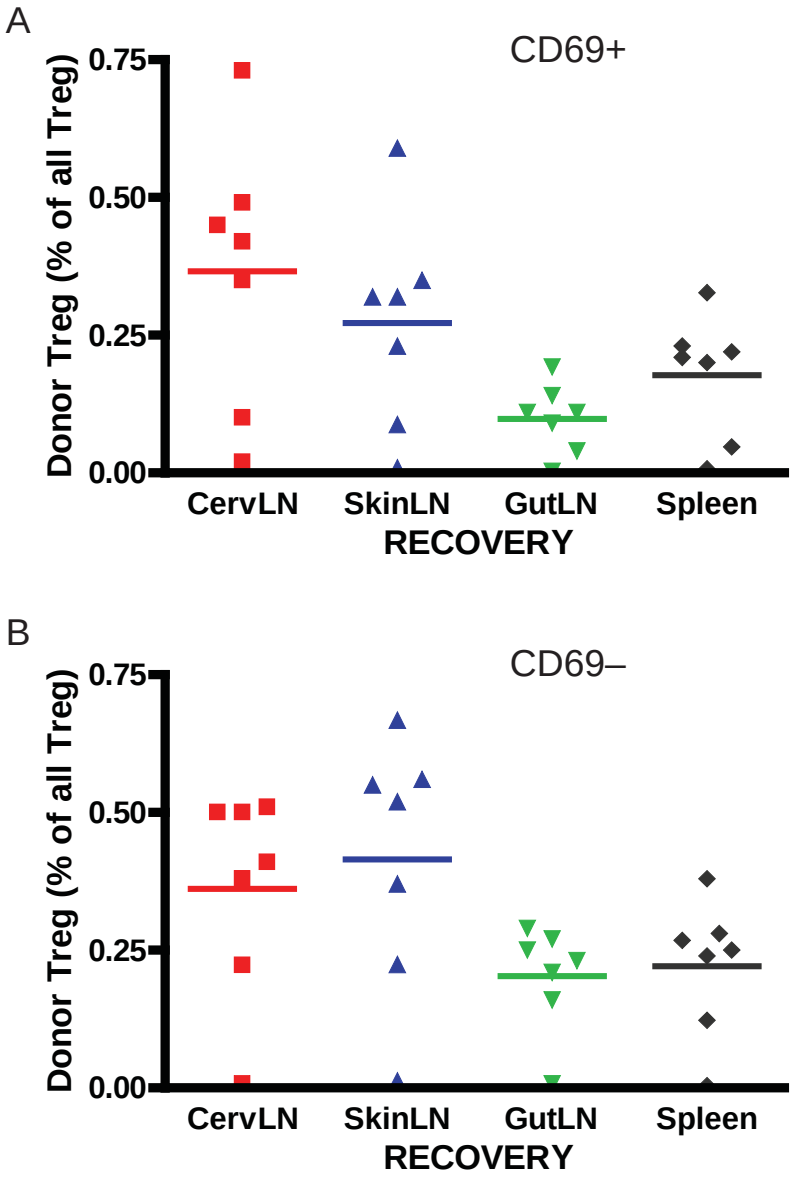
SUPPORTING INFORMATION FIGURE 2.



SUPPORTING INFORMATION FIGURE 3.

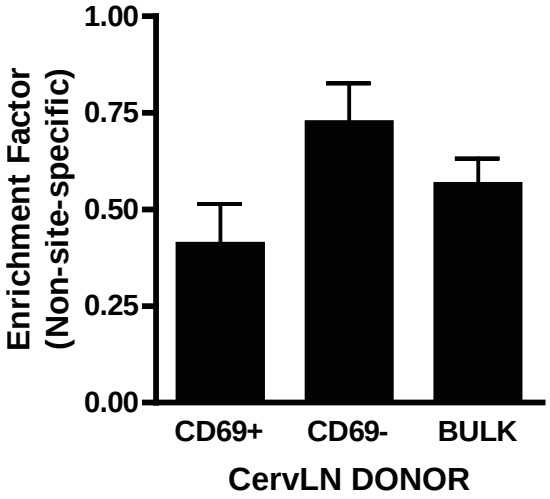


SUPPORTING INFORMATION FIGURE 4.



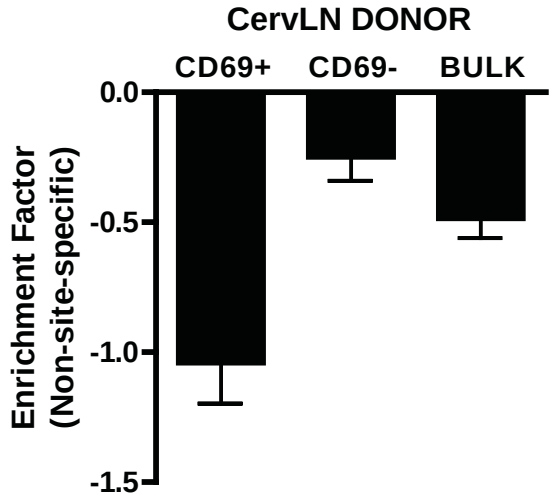
SUPPORTING INFORMATION FIGURE 5.

A



SkinLN Recovery

B



GutLN Recovery