Cascio, et al. Figure S1







## Cascio, et al. Figure S4



---- LP-Rag2<sup>-/-</sup>

LP-Rag2+/+

## SUPPLEMENTAL FIGURE LEGENDS

Figure S1. Bulk APCs from the SP and LP contain similar percentages of professional APC subsets. Bulk SP (A) and LP (B) APCs were harvested from C57BL/6 mice and stained with Abs to B220, CD19, PDCA-1, CD11b, CD11c, and F4/80 and analyzed by flow cytometry. The plots show a representative gating of the different APCs subsets. The bar graphs show the mean percentage  $\pm$  SE of B cells (B220<sup>+</sup>CD19<sup>+</sup>), pDC (CD11c<sup>+</sup>B220<sup>+</sup>pDCA-1<sup>+</sup>), conventional DC (CD11c<sup>+</sup>B220<sup>-</sup>pDCA-1<sup>-</sup>) and macrophages (CD11b<sup>+</sup>CD11c<sup>-</sup>F4/80<sup>+</sup>) among total APCs from three independent experiments.

Figure S2. Both antigen and T cells are required for lamina propria APCs to acquire tolerogenic function. Proliferative responses of 2D2 T cells were measured upon stimulation by MOG peptide-loaded LP APCs harvested from  $Rag2^{+/+}$  and  $Rag2^{-/-}$  C57BL/6 mice recipient of agg Ig-MOG + STI or agg Ig-W + STI. Data represents three independent experiments. \*p < 0.003.

Figure S3. Peptide carryover from orally fed Ig chimeras does not block MHC class II molecules of LP APCs. OT-II and 2D2 TCR transgenic mice were given oral agg Ig-OVA, agg Ig-MOG, or agg Ig-W along with STI and their LP were tested for presentation of unmatched peptide. The left panel shows proliferative responses of OVA-specific OTII T cells stimulated with OVA peptide presented by  $LP_{OTII/Ig-MOG}$  or  $LP_{OTII/Ig-W}$ . The right panel shows proliferative responses of MOG-specific 2D2 T cells stimulated with MOG peptide presented by  $LP_{2D2/Ig-OVA}$  or  $LP_{2D2/Ig-W}$ .

**Figure S4. In vivo exposure to oral Ig-MOG does not affect the expression of common costimulatory molecules on lamina propria APCs.** LP APCs were harvested from Rag2<sup>+/+</sup> and Rag2<sup>-/-</sup> C57BL/6 mice recipient of agg Ig-MOG + STI and the expression of co-stimulatory molecules was analyzed on specific subsets by flow cytometry. Representative histograms from at least three independent experiments are shown in comparison to isotype matched control (shaded).