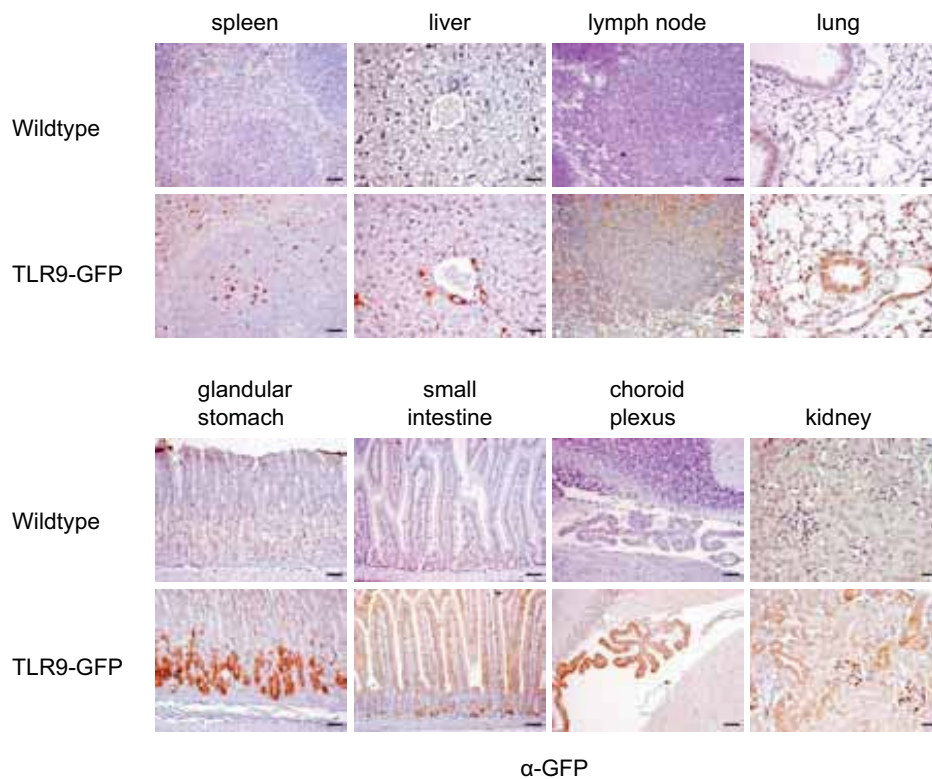


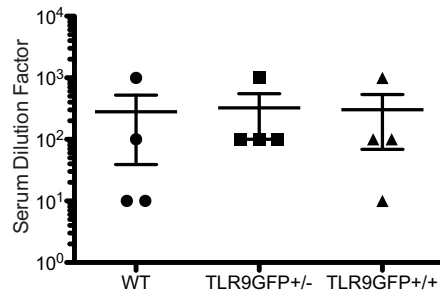
Supplementary Figure 1



Suppl. FIGURE 1. TLR9-GFP is expressed in different tissues of TLR9-GFP transgenic mice. Organs and tissue of 12-week-old mice heterozygous for transgenic TLR9-GFP (TLR9-GFP^{+/+}/WT) were formalin fixed, paraffin embedded and stained with an anti-GFP antibody. Data is representative of two TLR9-GFP^{+/+}/WT mice and two WT littermates. Scale bar = 25 μ m for lung and kidney and 50 μ m for all other organs.

Supplementary Figure 2

A



B

Table I

Lymphocyte populations in periphery (%)				
	WT	TLR9-GFP/WT	TLR9 KO	TLR9-GFP/KO
Spleen				
B220 ⁺ CD3 ⁺	2.2	1.7	1.7	1.5
B220 ⁺ CD23 ⁺	72.7	79.8	76.9	72.7
B220 ⁺ CD21 ⁺	10.6	6.4	6.4	9.3
CD4 ⁺	61.2	50.8	58.7	60.0
CD8 ⁺	28.6	36.6	28.7	21.5
!				
B cell subsets at different developmental stages (%) ^a				
Bone Marrow				
Pro B	13.6	20.7	16.1	23.0
Pre B	42.5	39.2	32.5	23.8
Immature B	40.3	36.6	47.8	51.1
Spleen				
Immature B	15.7	16.0	13.6	10.8
Mature B	84.2	83.9	86.3	89.1
!				
T cell subsets in thymus (%)				
Thymus				
CD4 ⁺	15.9	16.8	18.35	23.3
CD4 ⁺ CD8 ⁺	72.7	71.2	70.7	57.5
CD8 ⁺	2.5	3.1	3.1	3.7
CD4 ⁺ CD8 ⁻	1.3	2.2	1.7	4.0

^a In bone marrow, Pro B cells were scored based on B220⁺CD43⁺ expression. Pre B cells were CD43⁺B220⁺IgM⁻, and immature B cells were CD43⁺B220⁺IgM⁺. In spleen, immature B cells were B220⁺CD93⁺ and mature B cells were B220⁺CD93⁻. Values are averages of at least 3 mice except for bone marrow and thymus, with 1-2 mice per group.

!

Suppl. FIGURE 2. TLR9-GFP mice do not develop antinuclear antibodies and have normal lymphocyte development. (A) Sera from four of each 10-12 month old WT, TLR9-GFP mice with one (+/-) or two (++) copies of the TLR9-GFP transgene were extracted from the mouse, and serial dilutions of sera added to slides containing Hep-2 cells. Slides were washed and incubated with FITC-conjugated anti-Ig antibody and cells visualized with a fluorescence microscope. Serum dilution factor as quantification of levels of ANA was calculated at the dilution factor at which no signal could be detected. (B) T and B cell subsets were determined in spleen, bone marrow and thymus based on expression of surface markers.