Figure S1 Analysis of off-site target candidates at predicted sites in the Syrian hamster genome. A: PCR products flanking each predicted off-target (OT) site (#1–6) in WT and mutant founder hamster; B: PCR product sequencing confirmed no off-target cleavage. Representative sequencing analysis of founder is shown.
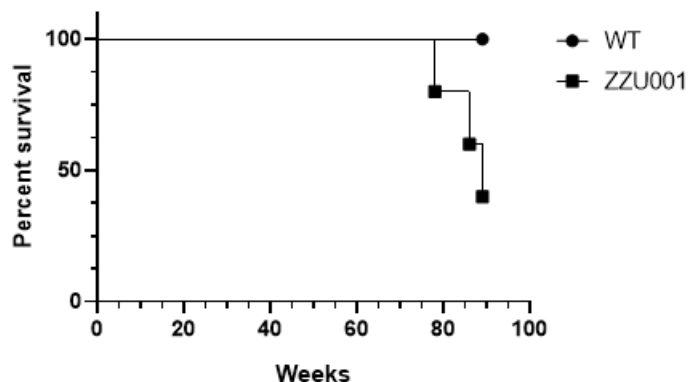
Figure S2 ZZU001 hamsters survival analysis. ZZU001 Syrian hamsters survive for over 72 weeks (1.5 years).   The median survival time exceeds 89 weeks (n=5/group).
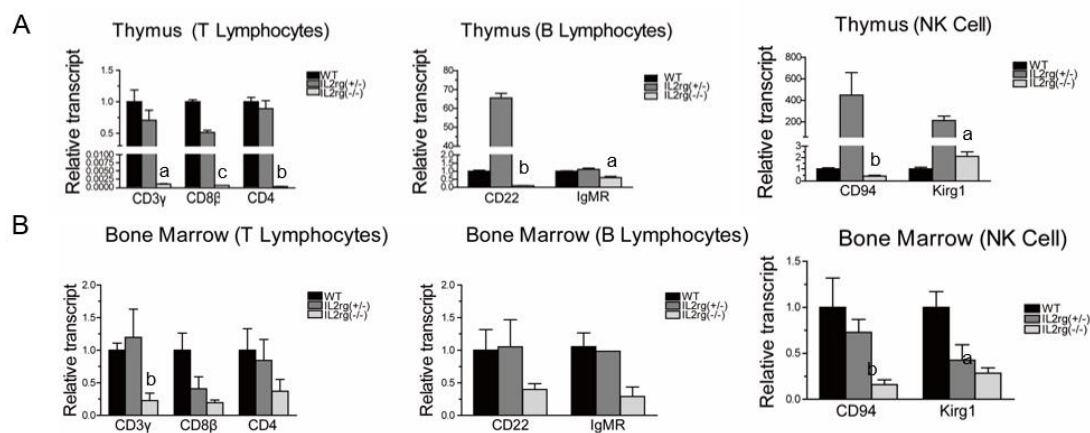

Figure S3 RT-qPCR analysis of lymphocyte-specific genes expression in thymus and bone marrow. A, B: Lower levels of mRNAs for T, B, NK lymphocyte-specific genes were detected. The amount of the indicated transcripts in thymus homogenates and bone marrow was determined by RT-qPCR using the $2^{\wedge}$ (-$\Delta\Delta$Ct) method. n=3. [a]$P<0.05$, [b]$P<0.01$, [c]$P<0.001$.
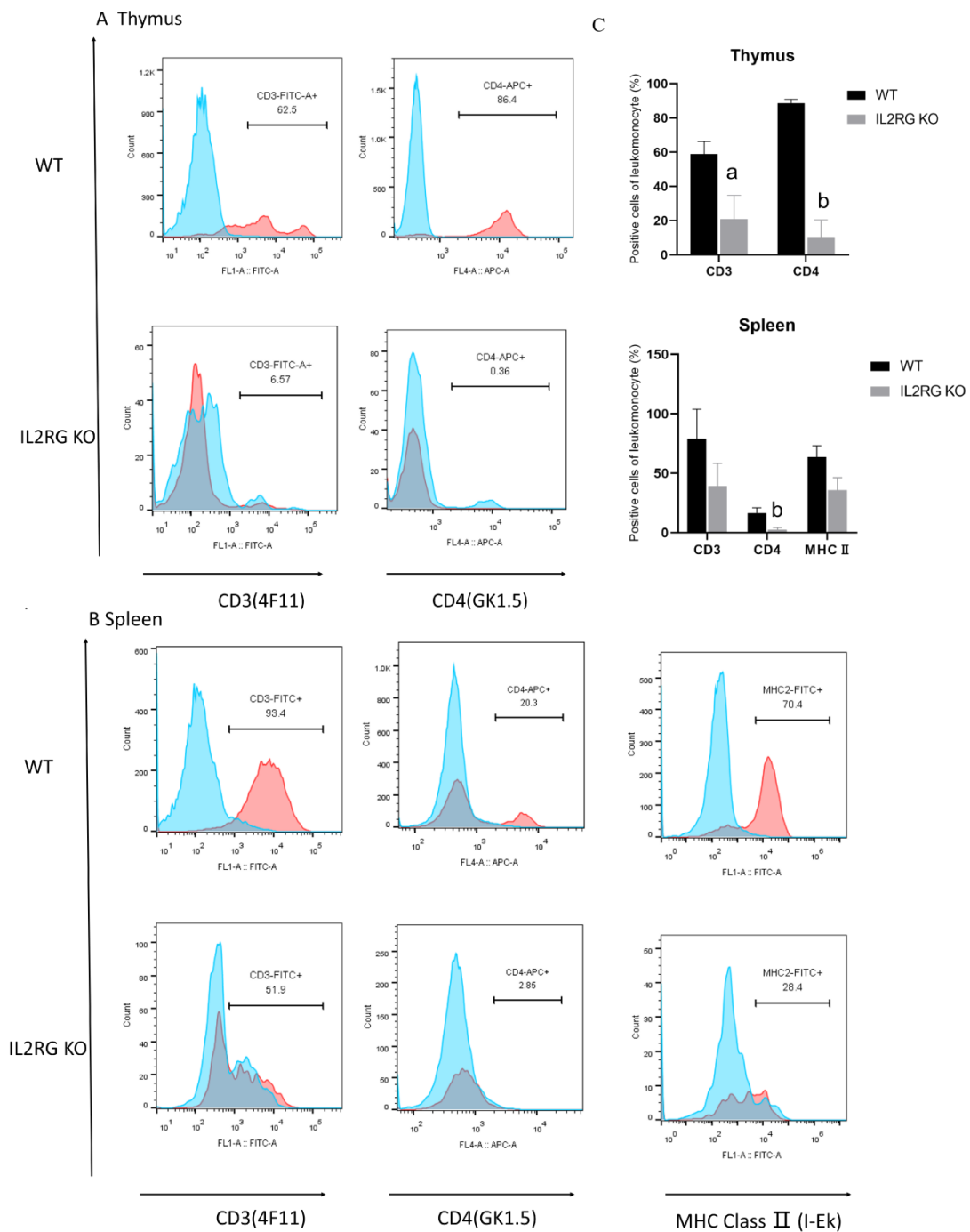
Figure S4 Flow cytometric analysis of cell populations isolated from thymus and spleen in WT and ZZU001 hamsters. A, B: CD3$^+$ and CD4$^+$ were absent from ZZU001 hamster thymus and spleen. MHC Class II cells were absent from ZZU001 hamster spleen. [a]$P<0.05$, [b]$P<0.01$. n=3.

Table S 1. List of PCR primers used in the study

Category	Name	Sequence (5`-3`)	Note
For designated target	GH IL2RG-F	GAGAGTGGTTCAGGGTTCTGACA	For sgRNA/Cas9-IL2RG
	GH IL2RG-R	TGGGCTGGAGCTCAGAACTG	For sgRNA/Cas9-IL2RG
For RT-qPCR	GH IL2RGqpcr-F	GAACCCTAGATTTTCCTTGCC	
	GH IL2RGqpcr-R	GTATGACTCCCCCAGTGAAC	
For potential off-site targets	OT1F	ACCCAGGACCTCAAGCATGC	
	OT1R	TGACCTGAGTTCGATTCCCC	
	OT2F	TGGAAGCAGTGACCAGGCTC	
	OT2R	GGGACCTCATTAGCCTGGAG	
	OT3F	CTCCAGATTCTAAGATTAGCAATGACA	
	OT3R	TAAGAGTGACTTGTGTGGCCTGAA	
	OT4F	GTCTGGCATCTAATTGCAAAGCA	
	OT4R	CAGAATGAATACTCAAGGAAATTGCG	
	OT5F	ATGATGGCATAAGATCTGGG	
	OT5R	ACTGGATCACAGGTTTTAGG	
	OT6F	GCTTCTGTTTTCTGGCCAAG	
	OT6R	GCATCGTATTCCTGGTTCAG	

Table S 2. Production of IL2RG targeted Syrian hamsters by PN injection

PN injected	Recipient	Delivery	Total pups	No. Positive pups (%)
181	7	2	8	3 (37.5)

Table S3. Top six potential off-targets by sgRNA/Cas9-IL2RG

	Off-targets Sequence (5`-3`)	PAM	Off-target Score	Sequence ID	Location of sites
OT 1	**T**AGCAGCTGAAG**C**AGTAAGA	GAG	3.9	NW_004801614.1	21825982 to 21826004
OT 2	GA**AG**AGCTG**C**AGGAGTAAGA	AGG	2.5	NW_004801763.1	2501450 to 2501472
OT 3	**C**A**C**CAG**G**TGAAGGAGTAAGA	TGG	1.8	NW_004801711.1	1215823 to 1215845
OT 4	GA**C**CAGC**CA**AAGGAGTAAGA	CAG	1.6	NW_004801606.1	24382163 to 24382185
OT 5	GA**TA**AGCTGA**G**GGAGTAAGA	GAG	1.6	NW_004801604.1	66170938 to 66170960
OT 6	G**G**GC**C**GCTGAA**T**GAGTAAGA	GGG	1.5	NW_004801654.1	5120437 to 5120459

Nucleotide mismatches between the target sequence and the potential off-target sequences are in red letters. Our scoring calculation follows the Zhang

[Lab's method](). Score is from 0-100. We include off-target matches for NAG and NGG since both have measurable efficiency.