Supporting Information

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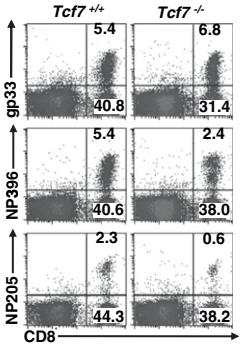

Fig. S1. Primary CD8 T cell response of *Tcf7*$^{-/-}$ mice to LCMV infection. *Tcf7*$^{+/+}$ (B6) and *Tcf7*$^{-/-}$ mice were infected with LCMV and the CD8 T cell response was investigated at day 8 postinfection using phycoerythrin-labeled D^b gp33 (KAVYNFATA), D^b NP396 (FQPQNGQFI), and K^b NP205 (YTVKYPNL) tetramers. Density plots show gated peripheral blood lymphocytes stained with the indicated tetramer and anti-CD8 mAb. Numbers indicate the percentage of cells in the respective quadrants.

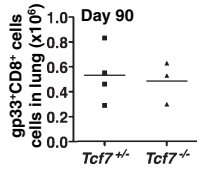

Fig. S2. Antigen-specific CD8 T cells in nonhematopoietic organs of *Tcf7*$^{-/-}$ mice. Symbols depict the absolute number of gp33-specific CD8 T cells in the lung of individual mice at day 90 post LCMV infection.

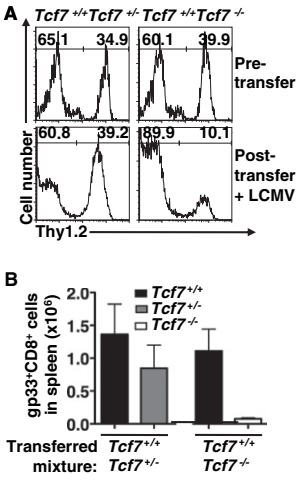

Fig. S3. Competitive recall response of *Tcf7*$^{-/-}$ and *Tcf7*$^{+/-}$ CD8 T cells to LCMV challenge infection. Splenocytes from LCMV immune *Tcf7*$^{+/-}$ or *Tcf7*$^{-/-}$ mice (Thy1.2 CD45.2) were mixed with those from *Tcf7*$^{+/+}$ (B6) mice (Thy1.1 CD45.2) (all around day 50 postinfection) such that each population contained an equal number (5×10^4) of gp33+ CD8+ T cells (*A* Upper). After transfer into naïve B6 recipients (Thy1.2 CD45.1), recipient mice were infected with LCMV and spleens were analyzed 5 days later (*Lower*). Histograms show gated CD45.2+ gp33$^+$ CD8$^+$ cells stained for Thy1.2. Numbers indicate the percentage of *Tcf7*$^{+/-}$ or *Tcf7*$^{-/-}$ (Thy1.2+) relative to *Tcf7*$^{+/+}$ (B6) (Thy1.2-) cells. (*B*) The bar graph depicts the mean number (± SD) of gp33-specific CD8 T cells derived from *Tcf7*$^{+/-}$ or *Tcf7*$^{-/-}$ (Thy1.2) or from cotransferred *Tcf7*$^{+/+}$ (B6) (Thy1.1) donors in spleens of recipient mice 5 days after LCMV challenge infection.

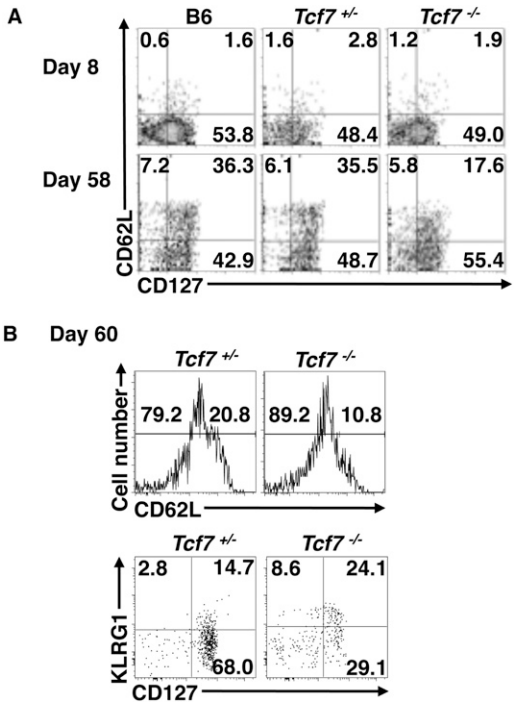

Fig. S4. Phenotypic analysis of LCMV-specific CD8 T cells (*A*) Density plots show the expression of CD127 (IL7Rα) versus CD62L in gated gp33-specific CD8 T cells from the indicated LCMV immune mice at day 8 and day 58 post-LCMV infection. (*B*) Histograms show CD62L expression among gated CD8 T cells of naive and of day 58 LCMV immune mice (*Top*) and among gated gp33-specific CD8 T cells (*Middle*). Dot plots show the expression of CD127 versus KLRG1 among gated CD62L^{hi} cells (*Bottom*). The respective gating is shown in the middle panel and was done according to the CD62L levels of naive CD8 T cells. Numbers indicate the percentage of cells in the respective gate or quadrants.

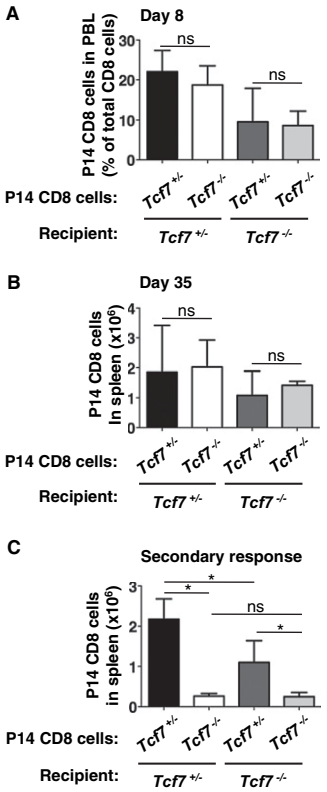

Fig. S5. Immune response of *Tcf7*$^{+/-}$ and *Tcf7*$^{-/-}$ P14 CD8 T cells adoptively transferred into *Tcf7*$^{+/-}$ and *Tcf7*$^{-/-}$ hosts. (*A*) Splenocytes from *Tcf7*$^{+/-}$ or *Tcf7*$^{-/-}$ mice containing 3×10^4 P14 CD8 T cells (CD45.2) were transferred into nontransgenic *Tcf7*$^{+/-}$ or *Tcf7*$^{-/-}$ recipients (CD45.1/CD45.2 heterozygous (CD45.1/2)) 1 day before LCMV infection. Bar graphs depict the mean percentage (± SD) of P14 CD8 cells in peripheral blood 8 days later. (*B*) Bar graphs depict the mean abundance (± SD) of P14 CD8 cells in recipient spleens at day 35 postinfection. (*C*) Splenocytes from primary recipients containing 3×10^4 P14 CD8 T cells (CD45.2) were transferred into secondary recipients (CD45.1/2) 1 day before LCMV challenge infection. Bar graphs depict the mean abundance (± SD) of P14 CD8 cells in spleens of secondary recipients 5 days later. (*) depicts significant difference ($P < 0.05$); ns, not significantly different ($P > 0.05$) based on Student's *t* test. The data confirm that KO cells transferred into WT hosts lack secondary expansion capacity, showing that Tcf-1 expression in CD8 T cells is essential for secondary replicative function. The reciprocal transfer of WT cells into KO host yielded an intermediate secondary response, suggesting that additional, CD8 T cell-independent defects in *Tcf7*$^{-/-}$ mice impact the recall response of CD8 T cells.

Table S1. CD8 and CD4 T cells in the spleen of *Tcf7*$^{-/-}$ and P14 *Tcf7*$^{-/-}$ mice

Strain	*n*	CD8 (×10^6)	CD44^{low} (% of CD8)	CD4 (×10^6)	CD44^{low} (% of CD4)
Tcf7$^{+/-}$	14	3.8 ± 1.6	69 ± 12	6.2 ± 2.5	77 ± 4
Tcf7$^{-/-}$	12	2.1 ± 1.0	47 ± 19	2.5 ± 1.0	51 ± 6
P14 Tcf7$^{+/-}$	4	7.0 ± 1.1	94 ± 1	nd	nd
P14 Tcf7$^{-/-}$	4	2.5 ± 0.7	75 ± 7	nd	nd

nd, not determined.

Table S2. Antigen-specific CD8 T cell responses to LCMV infection

	gp33		NP396		NP205	
Strain	Day 8	>Day 50	Day 8	>Day 50	Day 8	>Day 50
***Tcf7*$^{+/+}$**	12.0 ± 3.2*	9.8 ± 3.8	7.0 ± 4.7	1.0/3.0	3.5 ± 1.6	1.4 ± 0.5
***Tcf7*$^{+/-}$**	12.6 ± 4.7	7.0 ± 4.7	3.4 ± 1.1	1.6 ± 0.6	4.0 ± 2.1	1.0 ± 0.3
***Tcf7*$^{-/-}$**	11.9 ± 6.7	10.2 ± 7.1	5.1 ± 2.8	1.1 ± 0.4	2.5 ± 1.6	1.4 ± 0.8
	n > 17	*n* > 8	*n* = 3–7	*n* = 2–4	*n* = 3–7	*n* = 4

*Mean percentage (± SD) of gp33/D^b, NP396/D^b, or NP205/K^b tetramer binding cells among CD8 T cells in peripheral blood at day 8 and >day 50 post-LCMV infection. Corresponding stainings of naive mice yielded usually <0.2% of tetramer+ CD8 T cells.

Table S3. Antigen-specific CD4 T cell response to LCMV infection

	Day 8	>Day 50
Strain	IFNγ+CD4$^+$ ($\times 10^5$)	IFNγ+CD4$^+$ ($\times 10^5$)
Tcf7$^{+/+}$	2.8 ± 1.1*	0.37 ± 0.15
Tcf7$^{+/-}$	8.4 ± 6.5	0.29 ± 0.12
Tcf7$^{-/-}$	6.1 ± 2.5	0.47 ± 0.37

*Mean absolute number (± SD) of gp61-specific (IFNg+) CD4 T cells per spleen at day 8 and >day 50 post-LCMV infection. *n* = 3–4.