Figure S1. ***Genotypes of mice.*** Tail tips were collected from mice 3 weeks post-natally for genomic DNA extraction. Genotypes of all mice were examined by PCR assay using primers listed in (**A)** to detect the positivity or negativity for the *Cre* gene, *WT* and *fx* (floxed) alleles of the *Tak1* gene, *WT* and mutant alleles of *Tnfr1*, *Tnfr2* and *Fas* genes in all DNA samples. The gel pictures in (**B)** are examples of genotype data for *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$*r*$^{-}$*2*$^{-/-}$ and *Tak1*$^{-/-}$*Fas*$^{-/-}$ compound-mutant mice. **C.** BM MNCs were collected from *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$ mice on day 7 after polyI:C injection and seeded for CFU assay. The colonies were picked up individually for DNA extraction on day 10 of culturing. PCR was performed using primers TAK1 and 2 for *Fx* allele, and primers TAK3 and 4 for mutant allele. Lines 1 to 30 represent 30 different colony samples. Lines 31 and 32 are positive and negative controls, respectively.

Figure S2. ***Hematopoietic cell infiltration of liver in*** **Tak1**$^{-/-}$**Tnfr1**$^{-/-}$**r**$^{-}$**2**$^{-/-}$ ***mice***. Liver tissues were collected from *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$*r*$^{-}$*2*$^{-/-}$ mice 20 days after polyI:C injection. H & E staining (**A**) or TUNEL staining (**B**) of liver tissues from *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$*r*$^{-}$*2*$^{-/-}$ mice compared with *Tak1*$^{-/-}$ mice (8 days after polyI:C injection) and WT controls. Bar in figure is 100μm in length. **C**. Number of TUNEL-positive cells in each field of 400× microscopy was counted. Data in **C** are averages of 10 fields for each sample.

Figure S3. **A**. ***Colony forming ability of*** **Tnfr1**$^{-/-}$**, Tnfr2**$^{-/-}$**, Tnfr1**$^{-/-}$**r**$^{-}$**2**$^{-/-}$ ***and*** **Fas**$^{-/-}$ ***HSPCs are comparable to*** **WT** ***HSPCs.*** c-kit^{+} HSPCs were isolated from *Tak1*$^{fx/fx}$, *Tak1*$^{fx/fx}$*Tnfr1*$^{-/-}$, *Tak1*$^{fx/fx}$*Tnfr2*$^{-/-}$, *Tak1*$^{fx/fx}$*Tnfr1*$^{-/-}$*r*$^{-}$*2*$^{-/-}$ and *Tak1*$^{fx/fx}$*Tnfr2*$^{-/-}$ mice and infected with MSCV-GFP virus. Infected cells were purified by FACS to sort for GFP^{+} cells and seeded into methylcellulose medium for CFU assay. Exactly 30,000 GFP^{+} cells were sorted from each sample and placed into three dishes. Colonies were counted on day 10 of culturing. We found that comparable numbers of CFUs were observed in all samples. **B.** ***MSCV-Cre-GFP infection did not affect the colony forming ability of*** **Tak1**$^{fx/+}$ ***HSPCs.***

Figure S4. ***Receptors for several inflammatory factors are expressed in HSPCs and their levels are increased in*** **Tak1**$^{-/-}$**Tnfr1**$^{-/-}$ ***HSPCs.*** **A**. BM MNCs were collected from WT mice and stained with lineage cocktail and c-kit antibodies, as well as antibodies specific to receptors for inflammatory factors. Levels of receptors for inflammatory factors were compared among HSPCs (lin^{-}c-Kit^{+}), CPs (Lin^{low}c-kit^{+}) and differentiated cells (Lin^{+}). **B.** BM NMCs were collected from *Tnf1*$^{-/-}$ and *Tak1*$^{-/-}$*Tnf1*$^{-/-}$ mice on day 7 after three polyI:C injections (on days 1, 3, & 5). Levels of receptors for inflammatory factors were examined in HSCs (lin^{-}Sca1^{+}c-kit^{+}) and compared between the *Tnf1*$^{-/-}$ and *Tak1*$^{-/-}$*Tnf1*$^{-/-}$ mice.

Figure S5. ***TRAIL, IL-1β and IFN-α did not further enhance the death of*** **Tak1**$^{-/-}$**Tnfr1**$^{-/-}$ **r2**$^{-/-}$ ***HSPCs.*** c-kit^{+} HSPCs were insolated from *Tak1*$^{fx/fx}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ mice. These cells were infected with MSCV-Cre-GFP virus to induce *Tak1* deletion and cultured in HSPC medium with or without addition of inflammatory cytokines. MSCV-GFP infection was studied in parallel as a control. Dashed line represents the normalized data for MSCV-GFP infected HSPCs. Ratios of GFP^{+} cells (normalized against MSCV-GFP infection) were compared among different treatment groups.

Figure S6. ***Type-II apoptosis in*** **Tak1**$^{-/-}$ ***HSPCs.*** **A-B.** WT and *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ mice were injected with polyI:C on days 1 and 2 to induce *Tak1* deletion. BM MNCs were isolated on day 3 to analyze the mitochondrial membrane potential (Δψm) by JC-1 staining (**A**), as well as to examine the activity of caspases 9 and 3 by Western Blotting (**B**). **C**. c-Kit^{+} HSPCs were isolated from *Tak1*$^{fx/fx}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ mice and co-infected with MSCV-Cre-YFP and MSCV-Δ-caspase-9-GFP virus. Infected cells were purified by FACS (YFP^{+}GFP^{+} for co-infection) and incubated in HSPC medium for 24 hours. Death of infected cells was analyzed by Annexin-V staining. MSCV-Cre-YFP and MSCV-GFP infections were studied in parallel as controls. ****** indicates significantly reduced compared to Cre-only infection group.

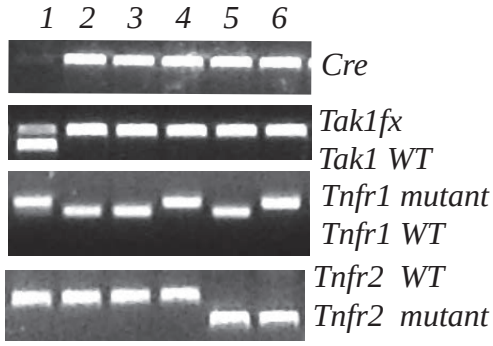
D & E. c-kit$^+$ HSPCs were collected from *Tak1*$^{fx/fx}$ (**D**) and *Tak1*$^{fx/fx}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ (**E**) mice and infected with MSCV-Cre-YFP virus or co-infected with MSCV-Cre-YFP and MSCV-test gene-GFP virus. MSCV-GFP infection was studied in parallel as a control. Infected cells were purified by FACS and incubated in HSPC medium for 24 hours to analyze cell death by Annexin-V staining, or were seeded into methylcellulose medium for CFU assay. ***** and ****** indicate significant difference compared to Cre-only infected group ($p<0.05$ and 0.01, respectively). Data shown in this figure are representative of 3 independent experiments.

Figure S7. ***ROS-induced cell death is not involved in apoptosis of HSPCs nor BMF in* Tak1$^{-/-}$ *mice.*** **A.** ROS levels were examined in WT and *Tak1*$^{-/-}$ HSPCs using dichlorodihydrofluorescein diacetate **(**CM-H_2DCFDA) staining followed by flow cytometric analysis**.** **B-C.** c-Kit$^+$ HSPCs were isolated from *Tak1*$^{fx/fx}$ and *Tak*$^{fx/fx}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ mice. These cells were infected with MSCV-Cre-GFP retrovirus to induce *Tak1* deletion *in vitro*. MSCV-GFP infections were studied in parallel as controls. Infected cells were cultured in HSPC medium with or without addition of NAC (N-acetyl-cysteine) or BHA (butylated hydroxyanisole) for 4 days with daily medium changes. Percentages of GFP$^+$ cells were examined at the indicated time points. The ratios of GFP$^+$ cells of MSCV-Cre-GFP-infected groups were normalized against the ratio in the MSCV-GFP-infected group. Dashed line represents the normalized data of MSCV-GFP-infected HSPCs. The reduction in the ratio of GFP$^+$ cells reflects the apoptotic loss of infected cells (*Tak1*-deleted cells). NAC and BHA treatments were unable to prevent this reduction in *Tak1*$^{-/-}$ (**B**) and in *Tak1*$^{-/-}$*Tnfr1*$^{-/-}$*r2*$^{-/-}$ (**C**) HSPCs. **D & E.** c-Kit$^+$ HSPCs were isolated from *Tak1*$^{fx/fx}$ mice. These cells were co-infected with MSCV-Cre-YFP (to induce *Tak1* deletion) and *SOD* (superoxide dismutase)-GFP or *Tak1*-GFP virus (to over-express SOD or Tak1). Co-infected cells were purified 16 hours after infection by sorting GFP$^+$YFP$^+$ cells and were seeded into methylcellulose medium for CFU analysis (**D**) or into HSPCs medium for apoptotic assay (**E**). MSCV-GFP and MSCV-Cre-YFP infections were studied in parallel as controls. *SOD* over-expression failed to prevent the apoptosis of *Tak1*$^{-/-}$ HSPCs. **F-I**. *Tak*$^{-/-}$ mice were injected with polyI:C on days 1, 3 and 5 to induce *Tak1* deletion, and were also treated with NAC on days 0 to 8 to reduce ROS levels. WT and additional *Tak1*$^{-/-}$ mice were injected with polyI:C only or NAC only to serve as controls. Femurs and spleens of the mice were collected on day 8 after induction of *Tak1* gene deletion. H & E-stained BM sections from the indicated genotypes of mice with or without NAC treatment are shown **(F)**. Numbers of total nucleated cells from two hind legs of each mouse were counted (**G**). Percentages (**H**) and absolute numbers (**I**) of HSCs and CPs were studied by flow cytometry. Data shown in **H & I** are an average of 5 mice from each genotype.

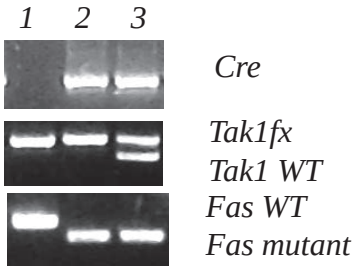
Figure S8. ***Apoptosis of hepatocytes in* Tak1$^{-/-}$ *mice is substantially prevented by antioxidant treatment***. *Tak1*$^{-/-}$ and littermate control mice (1 month old) were injected 3 times with polyI:C to induce *Tak1* gene deletion. Three *Tak1*$^{-/-}$ mice were also injected with an antioxidant (NAC) every day beginning one day prior to the first polyI:C injection. Livers of these mice were collected on day 8 after the injections. **A**. H & E-stained liver sections from indicated genotypes of mice. **B**. Apoptosis in liver sections was examined by TUNEL staining and the number of TUNEL positive cells in each field of 400× microscopy was counted. Data in **C** are averages of 10 fields for each sample.

A.	Primers	Products
Cre Forward	TACCTGGAAAATGCTTCTGT	810bp
Cre Reverse	TGATCTCCGGTATTGAAACT	
Tnfrsf1a-1	GGATTGTCACGGTGCCGTTGAAG	WT = 120 bp
Tnfrsf1a-2	TGACAAGGACACGGTGTGTGGC	MUT = 155 bp
Tnfrsf1a-3	TGCTGATGGGGATACATCCATC	
Tnfrsf1a-4	CCGGTGGATGTGGAATGTGTG	
Tnfrsf1b-1	CCGGTGGATGTGGAATGTGTG	MUT = 160 bp
Tnfrsf1b-2	AGAGCTCCAGGCACAAGGGC	WT = 257 bp
Tnfrsf1b-3	AACGGGCCAGACCTCGGGT	
Fas-1	TAGAAAGGTGCACGGGTGTG	MUT=217 bp
Fas-2	GTAAATAATTGTGCTTCGTCAG	WT=179 bp
Fas-3	CAAATCTAGGCATTAACAGTG	
TAK1	GGCTTTCATTGTGGAGGTAAGCTGAGA	flox 320bp
TAK2	GGAACCCGTGGATAAGTGCACTTGAAT	WT 280bp
TAK3	GCAACTTCGACAACTTGCCTTCCTGTG	mutant 1000bp
TAK4	GCACTTGAATTAGCGGCCGCAAGCTTATAACT	

B.



1. *Mx1Cre-Tak1fx/+Tnfr1-/-r2+/+*
2. *Mx1Cre+Tak1fx/fxTnfr1+/+r2+/+*
3. *Mx1Cre+Tak1fx/fxTnfr1+/+r2+/+*
4. *Mx1Cre+Tak1fx/fxTnfr1-/-r2+/+*
5. *Mx1Cre+Tak1fx/fxTnfr1+/+r2-/-*
6. *Mx1Cre+Tak1fx/fxTnfr1-/-r2-/-*



1. *Tak1fx/fxfas+/+*
2. *Mx1Cre+Tak1fx/fxFas-/-*
3. *Mx1Cre+Tak1fx/+Fas-/-*

C.

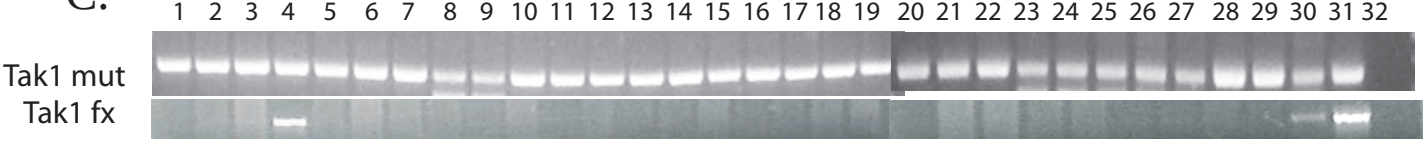


Figure S1

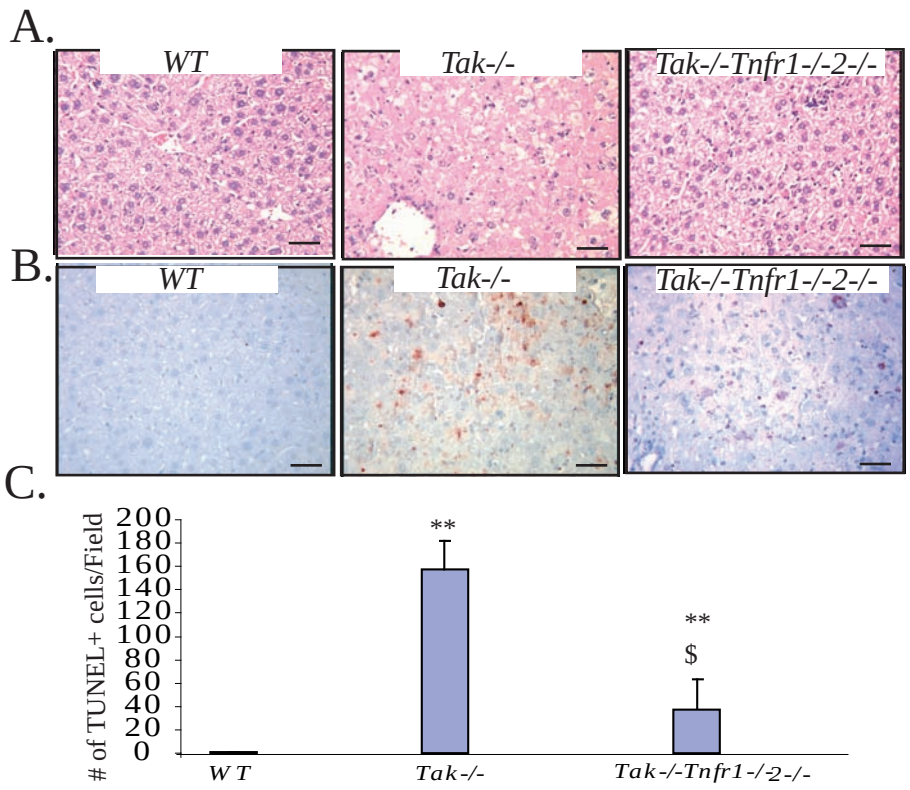
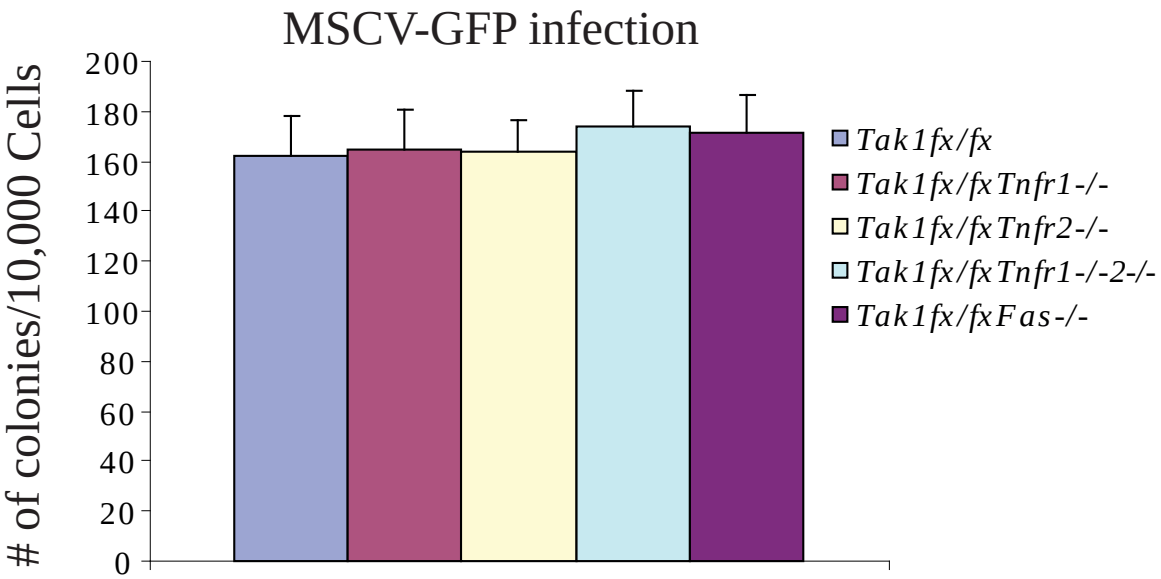

A.
WT
Tak-/-
Tak-/-Tnfr1-/-2-/-
B.
WT
Tak-/-
Tak-/-Tnfr1-/-2-/-
C.
# of TUNEL+ cells/Field
200
180
160
140
120
100
80
60
40
20
0
**
**
$
WT
Tak-/-
Tak-/-Tnfr1-/-2-/-

Figure S2

A.


MSCV-GFP infection
# of colonies/10,000 Cells
200
180
160
140
120
100
80
60
40
20
0
Tak1fx/fx
Tak1fx/fxTnfr1-/-
Tak1fx/fxTnfr2-/-
Tak1fx/fxTnfr1-/-2-/-
Tak1fx/fxFas-/-

B.

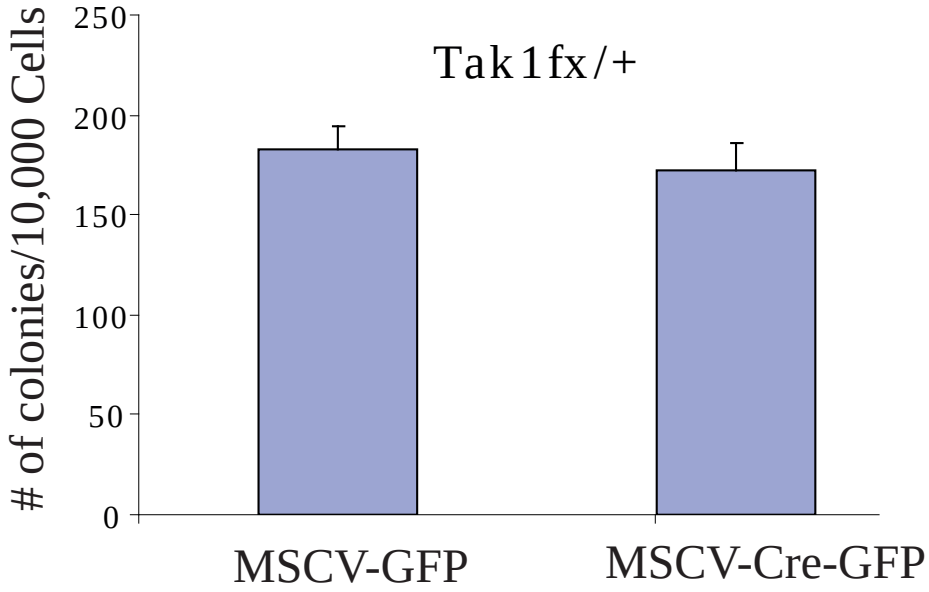

Tak1fx/+
# of colonies/10,000 Cells
250
200
150
100
50
0
MSCV-GFP
MSCV-Cre-GFP

Figure S3

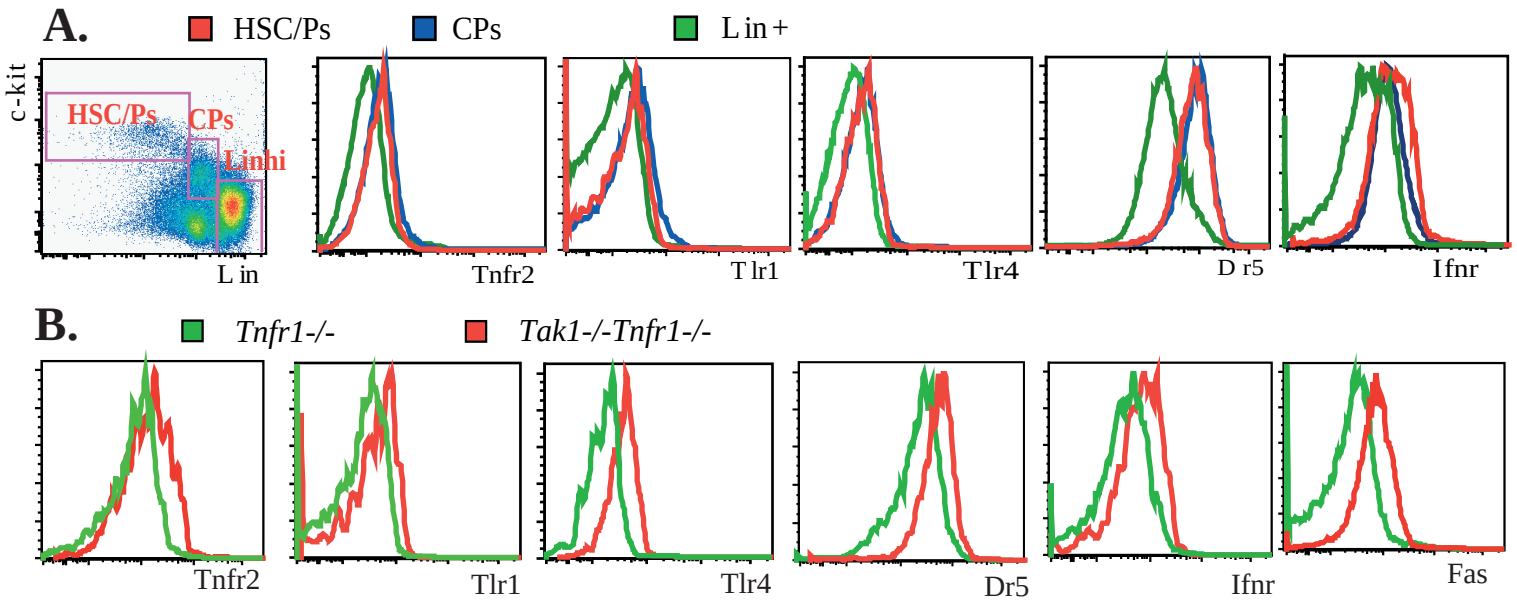
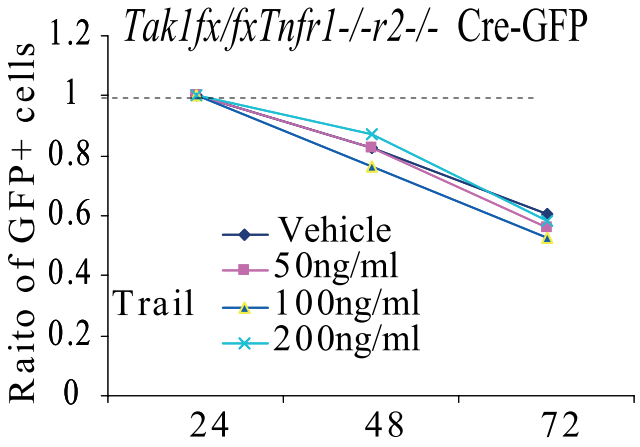
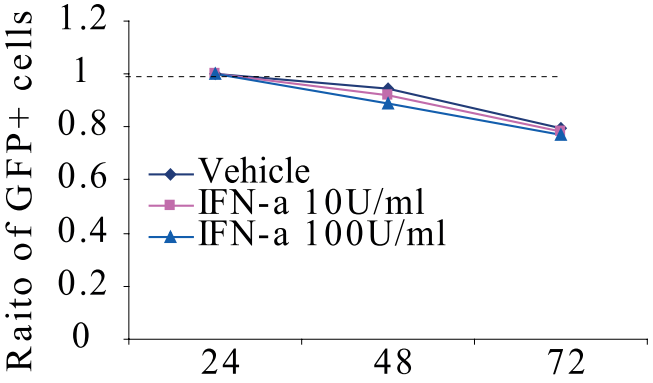

A.
HSC/Ps
CPs
Lin+
c-kit
HSC/Ps
CPs
Linhi
Lin
Tnfr2
Tlr1
Tlr4
Dr5
Ifnr
B.
Tnfr1-/-
Tak1-/-Tnfr1-/-
Tnfr2
Tlr1
Tlr4
Dr5
Ifnr
Fas

Figure S4

A.



B.



C.

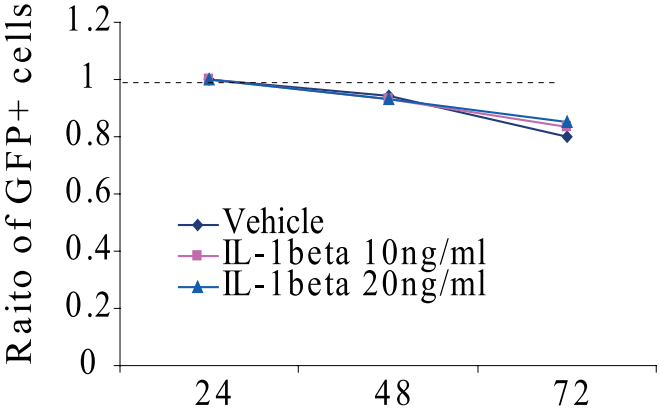


Figure S5

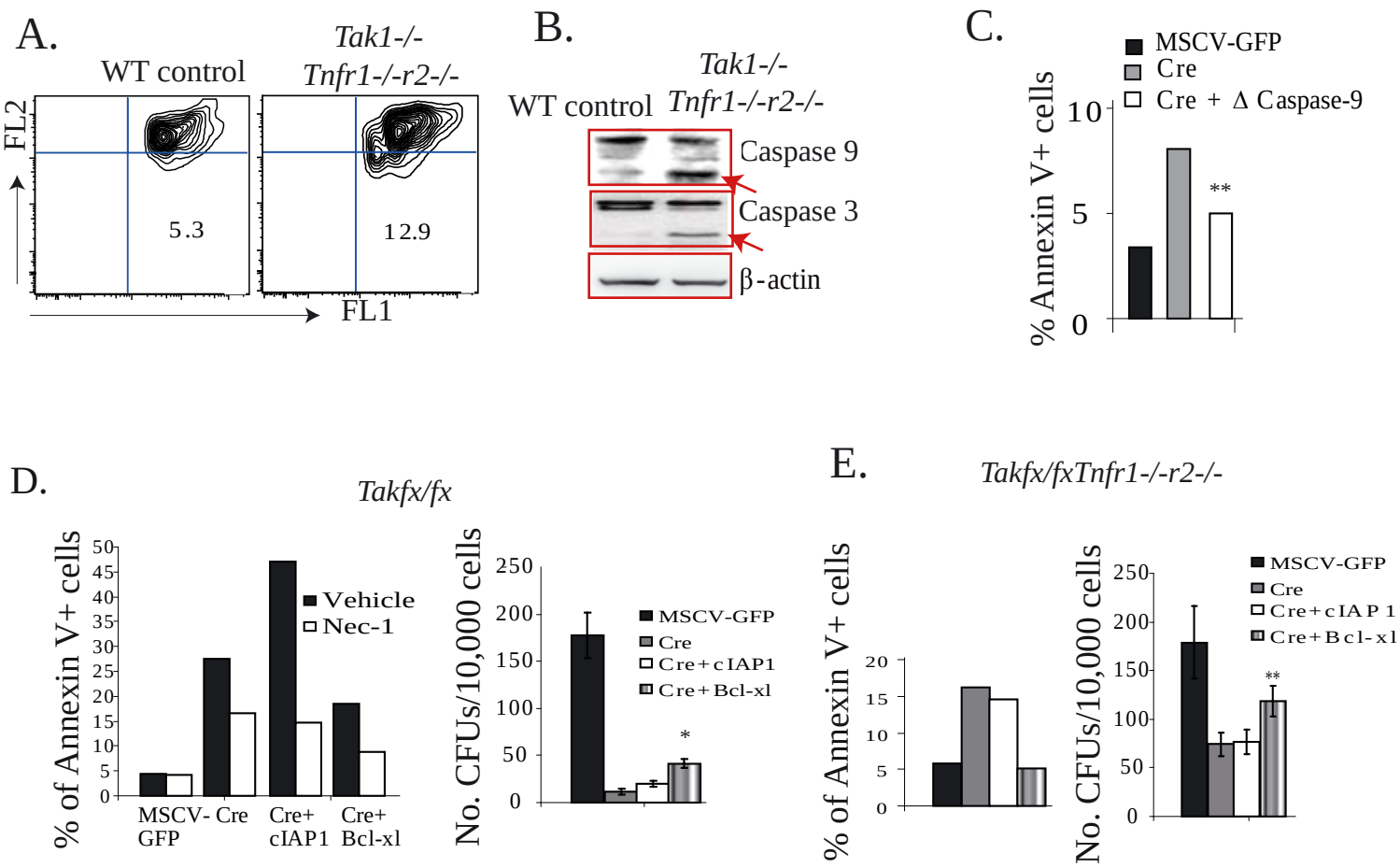

A.
WT control
Tak1-/-
Tnfr1-/-r2-/-
FL2
FL1
5.3
12.9
B.
WT control
Tak1-/-
Tnfr1-/-r2-/-
Caspase 9
Caspase 3
β-actin
C.
MSCV-GFP
Cre
Cre + Δ Caspase-9
% Annexin V+ cells
10
5
0
**
D.
Takfx/fx
% of Annexin V+ cells
50
45
40
35
30
25
20
15
10
5
0
Vehicle
Nec-1
MSCV-GFP
Cre
Cre+cIAP1
Cre+Bcl-xl
No. CFUs/10,000 cells
250
200
150
100
50
0
*
E.
Takfx/fxTnfr1-/-r2-/-
% of Annexin V+ cells
20
15
10
5
0
MSCV-GFP
Cre
Cre+cIAP1
Cre+Bcl-xl
No. CFUs/10,000 cells
250
200
150
100
50
0
**

Figure S6

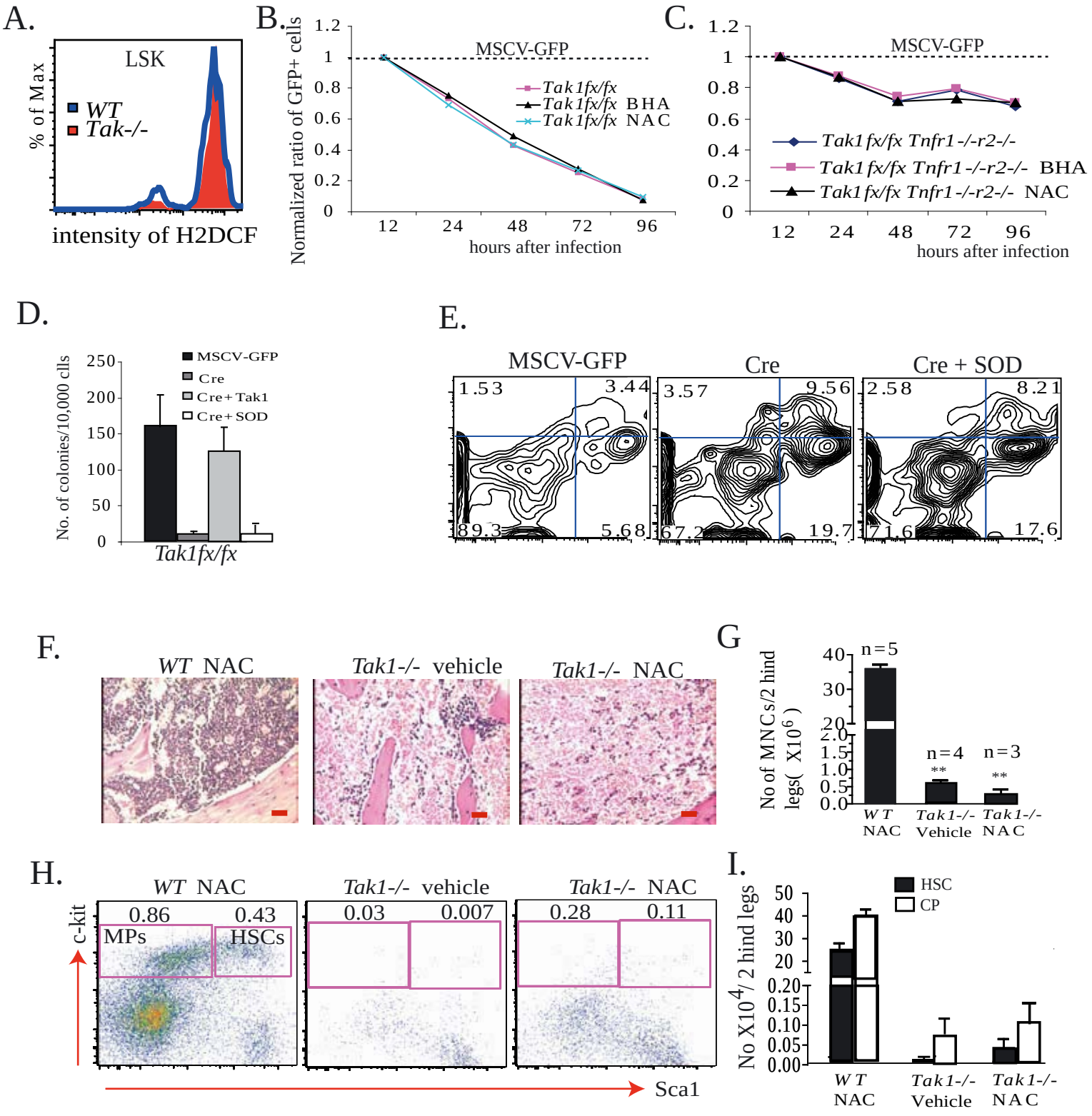

A.
LSK
% of Max
WT
Tak-/-
intensity of H2DCF
B.
MSCV-GFP
Tak1fx/fx
Tak1fx/fx BHA
Tak1fx/fx NAC
Normalized ratio of GFP+ cells
hours after infection
C.
MSCV-GFP
Tak1fx/fx Tnfr1-/-r2-/-
Tak1fx/fx Tnfr1-/-r2-/- BHA
Tak1fx/fx Tnfr1-/-r2-/- NAC
hours after infection
D.
MSCV-GFP
Cre
Cre+Tak1
Cre+SOD
No. of colonies/10,000 clls
Tak1fx/fx
E.
MSCV-GFP
Cre
Cre + SOD
1.53
3.44
89.3
5.68
3.57
9.56
67.2
19.7
2.58
8.21
71.6
17.6
F.
WT NAC
Tak1-/- vehicle
Tak1-/- NAC
G
No of MNCs/2 hind legs(X10^6)
n=5
n=4
n=3
**
**
WT NAC
Tak1-/- Vehicle
Tak1-/- NAC
H.
WT NAC
Tak1-/- vehicle
Tak1-/- NAC
0.86
0.43
MPs
HSCs
0.03
0.007
0.28
0.11
c-kit
Sca1
I.
HSC
CP
No X10^4/ 2 hind legs
WT NAC
Tak1-/- Vehicle
Tak1-/- NAC

Figure S7

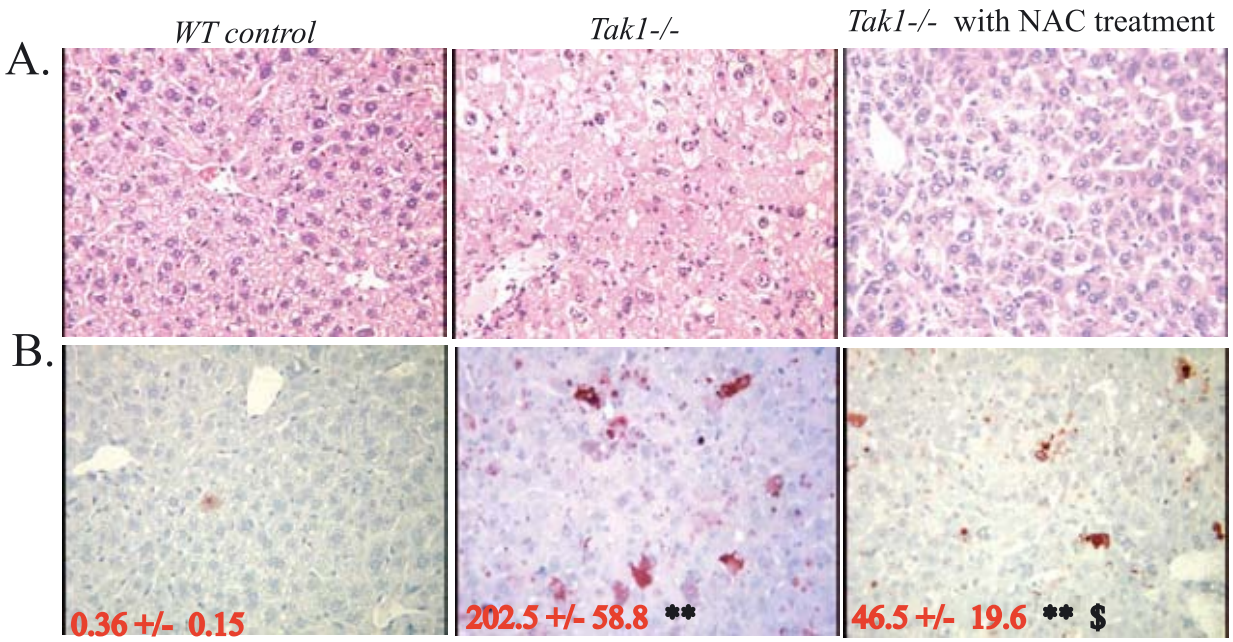

WT control
Tak1-/-
Tak1-/- with NAC treatment
A.
B.
0.36 +/- 0.15
202.5 +/- 58.8 **
46.5 +/- 19.6 ** $

Figure S8