Supporting Information

Kupz et al. 10.1073/pnas.1222047110

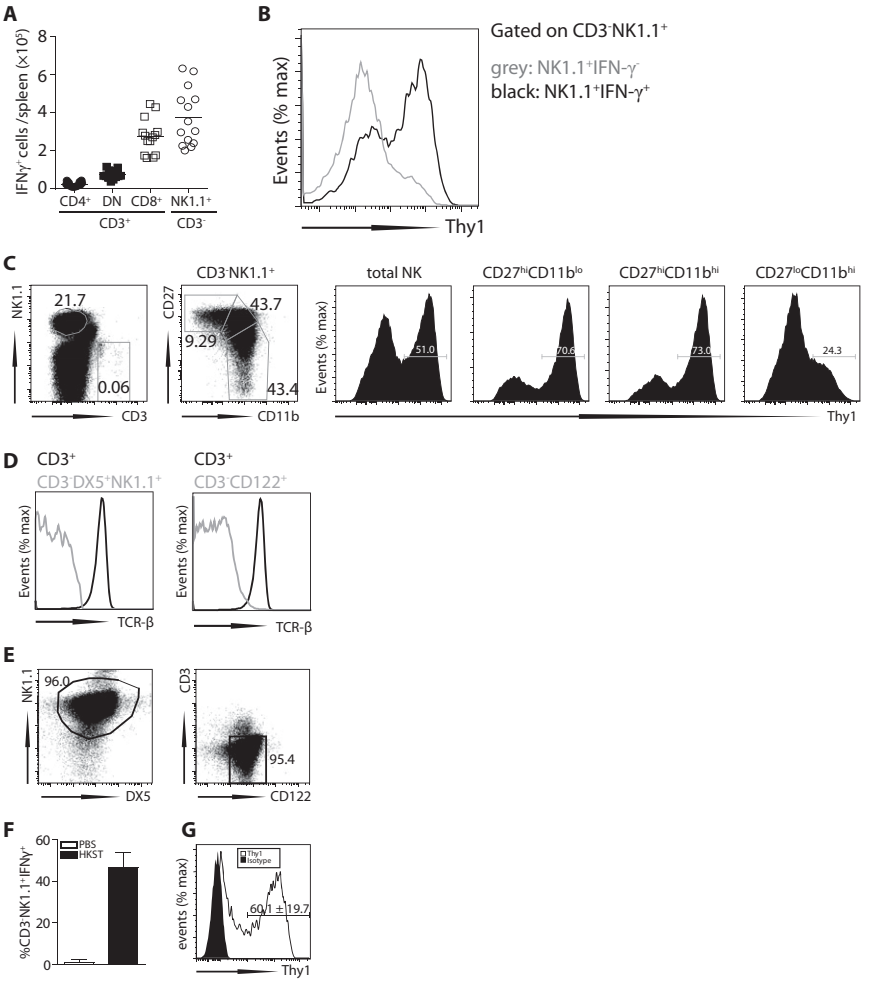

Fig. S1. Thy1-expressing natural killer (NK) cells are not T-cell contaminants. (*A* and *B*) Naïve B6 mice were i.v. injected with 1×10^8 cfu heat-killed *Salmonella enterica* serovar Typhimurium (HKST). IFN-γ secretion by lymphocyte subsets was assessed 2 h later in the spleen. IFN-γ^+ and IFN-γ^- NK 1.1^+ cells were assessed for expression of Thy1 (*B*). Total number of IFN-γ^+ cells among total $CD3^+$ and $CD3^-$ cells (*A*) and a representative histogram (*B*) are shown. (*C*) Viable single splenocytes from *Rag1/Je*$^{-/-}$ mice were assessed for expression of Thy1 on different NK cells subsets ($CD3^-$ $NK1.1^+$) defined by the expression of CD27 and CD11b. (*D*) Expression of T-cell receptor (TCR)-β on splenic $CD3^-DX5^+NK1.1^+$ (*Left*) and $CD19^-CD3^-CD4^-CD122^+$ (*Right*) cells in B6 mice in comparison with $CD3^+$ cells. (*E*–*G*) In vitro–activated NK cells from B6 mice were assessed for expression of NK1.1, DX5, CD122, and CD3 (*E*), the capacity to produce IFN-γ in vivo after transfer of two times 1×10^6 NK cells into naïve congenic recipients (*F*), and expression of Thy1 (*G*). Individual data points from at least three experiments (*A*) and representative FACS plots (*C* and *E*) and histograms (*B*–*D* and *G*) from at least two independent experiments are shown.

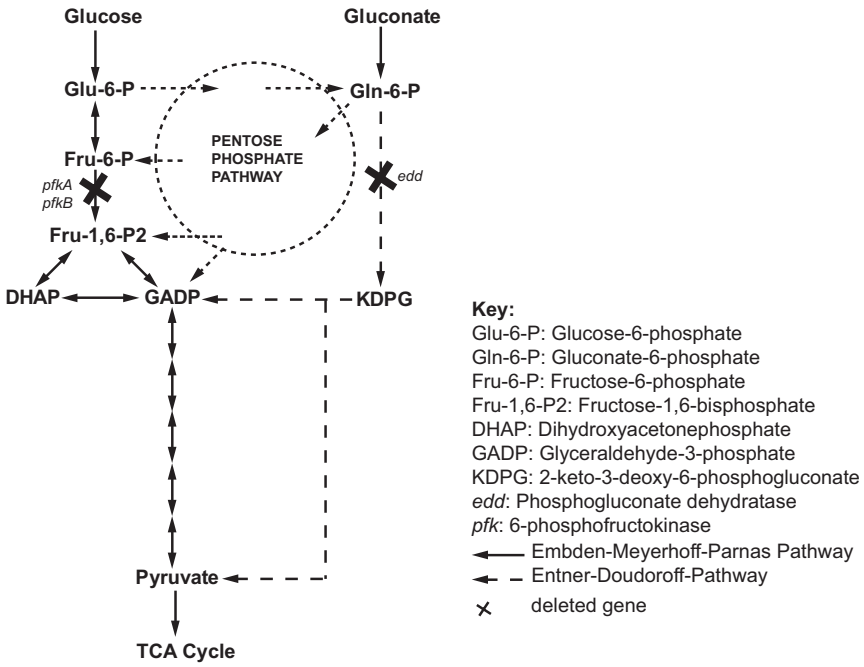

Fig. S2. Schematic presentation of mutation sites in SL1344 *Δedd ΔpfkA ΔpfkB*. Schematic representation of the central carbon metabolism. Sites of mutations to generate SL1344 *Δedd ΔpfkA ΔpfkB* are marked with a black cross.

Table S1. Overview of mouse strains used in this study

Mouse strain	Purpose
C57BL/6	WT
CD45.1 (Ly5.1)	Congenic cell marker
GK1.5Tg (1)	Transgenic expression of anti-GK1.5; $CD4^+$ T cell deficient
GK1.5/2.43Tg (2)	Transgenic expression of anti-GK1.5 and anti-2.43; $CD4^+$ and $CD8^+$ T cell deficient
Rag1/Je$^{-/-}$ (3)	Deletion of Rag1; T and B cell deficient
Rag2$^{-/-}\gamma c^{-/-}$ (4)	Deletion of Rag2 and common γ chain; lymphocyte deficient; NK cell recipient
IFN-γ$^{-/-}$ (5)	Deletion of IFN-γ; highly susceptible to *STM* infection
CD1d$^{-/-}$ (6)	Deletion of CD1d; NKT cell deficient
μMT (7)	Disruption of Ig μ chain; B cell deficient
ROR-γt$^{-/-}$ (8)	Deletion of RORγt; innate lymphoid cell and Th17 deficient

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2. Han WR, et al. (2000) Prolonged allograft survival in anti-CD4 antibody transgenic mice: Lack of residual helper T cells compared with other CD4-deficient mice. *Transplantation* 70(1): 168–174.
3. Mombaerts P, et al. (1992) RAG-1-deficient mice have no mature B and T lymphocytes. *Cell* 68(5):869–877.
4. Mazurier F, et al. (1999) A novel immunodeficient mouse model–RAG2 3 X common cytokine receptor γ chain double mutants requiring exogenous cytokine administration for human hematopoietic stem cell engraftment. *J IFN Cytokine Res* 19(5):533–541.
5. Dalton DK, et al. (1993) Multiple defects of immune cell function in mice with disrupted IFN-γ genes. *Science* 259(5102):1739–1742.
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7. Kitamura D, Roes J, Kühn R, Rajewsky K (1991) A B-cell-deficient mouse by targeted disruption of the membrane exon of the Ig mu chain gene. *Nature* 350(6317):423–426.
8. Eberl G, et al. (2004) An essential function for the nuclear receptor RORγt in the generation of fetal lymphoid tissue inducer cells. *Nat Immunol* 5(1):64–73.

Table S2. Monoclonal antibodies used for in vivo cell and cytokine depletion

Antibody	Clone	Injection dose per mouse	Source
Anti-CD8	2.43	250 μg initially then 200 μg twice weekly	WEHI
Anti-CD4	GK1.5	250 μg initially then 200 μg twice weekly	WEHI
Anti-Thy1.2	30-H12	250 μg initially then 200 μg twice weekly	WEHI
Anti-IFN-γ	HB-170–15	200 μg weekly	WEHI
Anti-NK1.1	PK136	250 μg initially then 200 μg twice weekly	WEHI

WEHI, Walter and Eliza Hall Institute of Medical Research, Melbourne, Australia.

Table S3. Oligonucleotides used to generate *Salmonella* Typhimurium SL1344 *Δedd ΔpfkA ΔpfkB*

Function	Primer name	Sequence (5′ to 3′)*
kan amplification	kanF	GTGTAGGCTGGAGCTGCTTC
	kanR	CATATGAATATCCTCCTTAG
edd deletion	edd2(ISceI)F	TAGGGATAACAGGGTAATCATCGGAATTCTTCTCTCGC
	edd2KanR	**GAAGCAGCTCCAGCCTACACA**GTAATGAAGACGTCTGCGGTAC
	edd2KanF	**CTAAGGAGGATATTCATATG**GGTTTACCATGCGTTTCATC
	edaedd(ISceI)R	TAGGGATAACAGGGTAATGAACAAATTGACGATTCGCCTGC
pfkA deletion	pfkA(ISceI)F	TAGGGATAACAGGGTAATGGTGCAGTCATTATTGGATCG
	pfkAKanR	**GAAGCAGCTCCAGCCTACA**CAGACTACCTCTGAACTTTGGAATGC
	pfkAKanF	**CTAAGGAGGATATTCATATG**ACATCATCGATGCGATTG
	pfkA(ISceI)R	TAGGGATAACAGGGTAATCGTCACGACATCGGCTTC
pfkB deletion	pfkB(ISceI)F	TAGGGATAACAGGGTAATGTCCATACCAGGTCATCG
	pfkBKanR	**GAAGCAGCTCCAGCCTACACA**CGTTACCTCCTGTTAGGCTG
	pfkBKanF	**CTAAGGAGGATATTCATATG**TGTTCTCGTGACGATACC
	pfkB(ISceI)R	TAGGGATAACAGGGTAATGGAATGGCACTTATTGTGC

*I-SceI restriction sites are underlined; *kan*-specific sequences are bold.