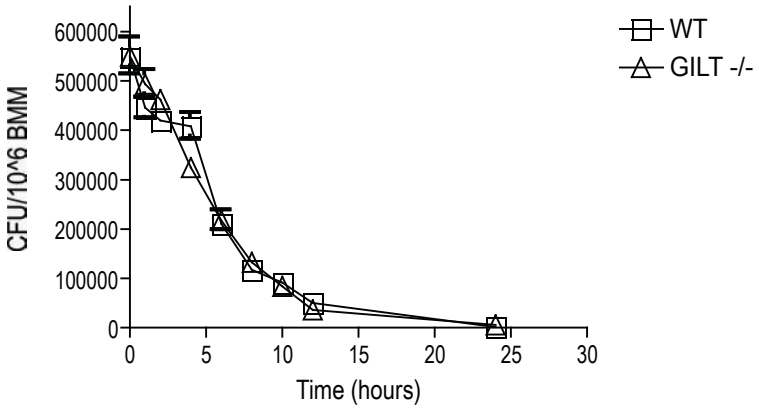
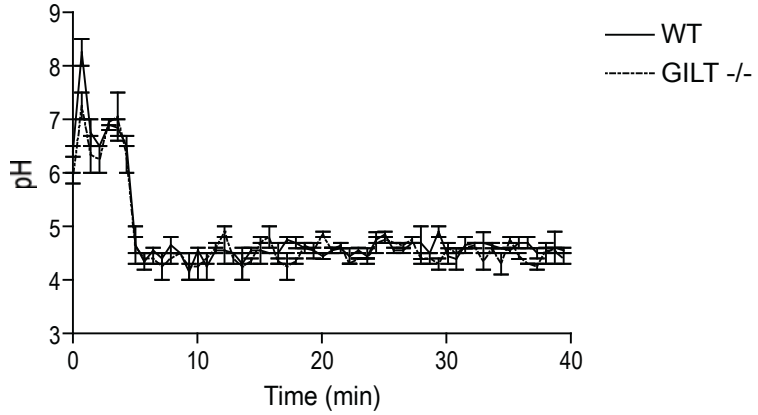


Supplemental Figure 1

a



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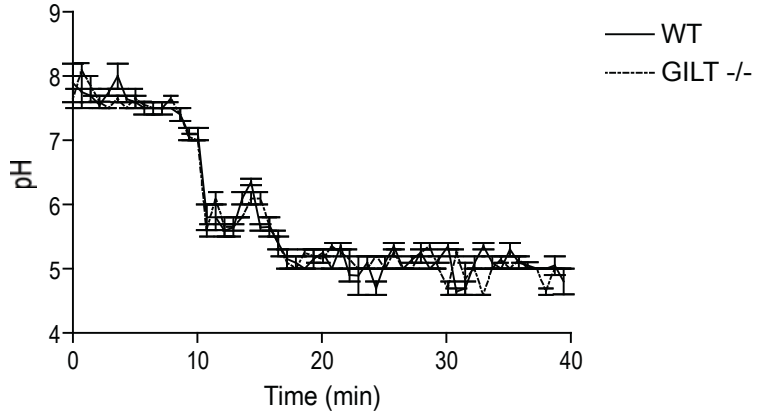
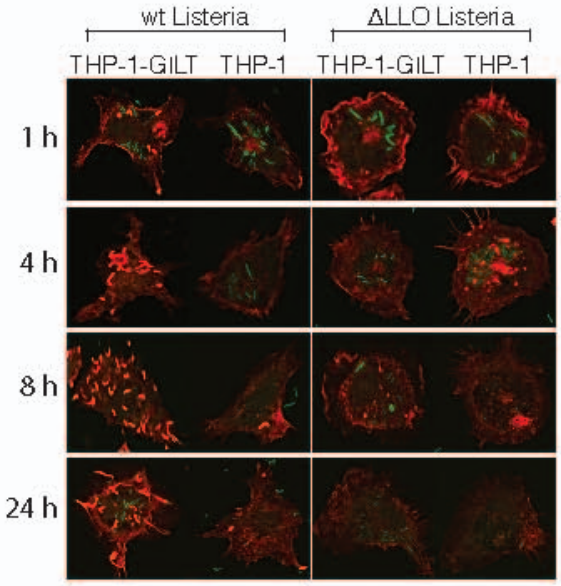
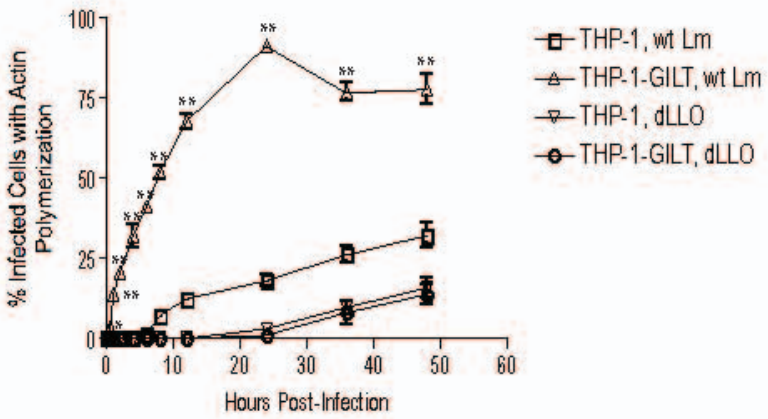


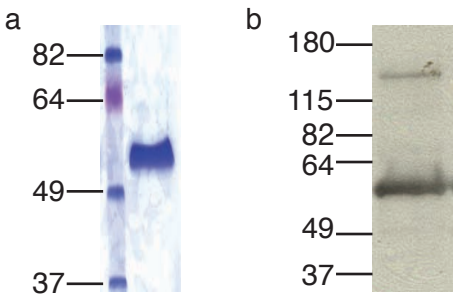
Supplemental Figure 2

a


wt Listeria
ΔLLO Listeria
THP-1-GILT
THP-1
THP-1-GILT
THP-1
1 h
4 h
8 h
24 h

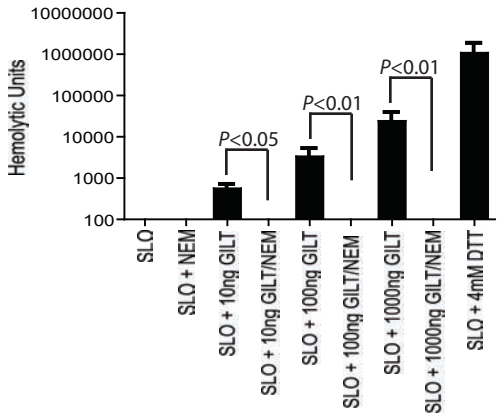
b


% Infected Cells with Actin Polymerization
Hours Post-Infection
THP-1, wt Lm
THP-1-GILT, wt Lm
THP-1, dLLO
THP-1-GILT, dLLO

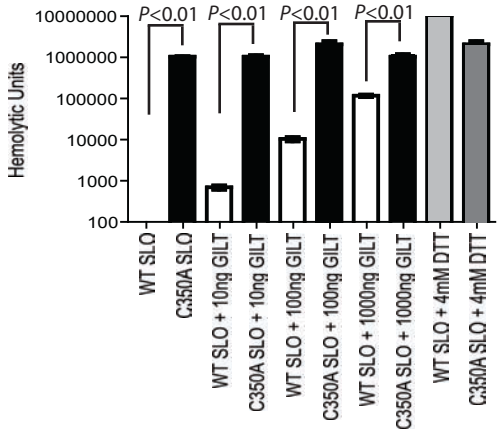


Supplemental Figure 4

a



b



Supplemental Figure 5

Supplemental Figure 1. In vitro killing of *E. coli* by wild type or GILT -/- bone marrow derived macrophages. Bone marrow-derived macrophages were infected with *E. coli* at an MO1 of 1 and cultured for the time points shown. At each time point, the macrophages were lysed and plated on bacterial plates. Colonies were counted the next day to determine CFUs. An average of three independent experiments is shown.

Supplemental Figure 2. Acidification of both the lysosome and the phagosome is the same in both wild type and GILT -/- bone marrow derived macrophages. Macrophages were incubated with indicator dextrans or beads (see Methods) on ice for 45 min. and then washed extensively to eliminate loosely bound dextran or beads. Cells were then plated in 96-well plates and fluorescence at alternating excitations of 450 and 490nm with emission at 520nm over a 40 min. period was read using a plate reader. The pH was calculated based on a standard curve from the ratio of the pH-insensitive excitation at 450nm and pH sensitive excitation at 490 nm. a, lysosomal pH. b, phagosomal pH. For both, an average of four independent experiments is shown.

Supplemental Figure 3. Phagosomal escape of *L. monocytogenes* is enhanced in THP-1 cells that constitutively express GILT. **a**, Actin polymerization in wild type THP-1 cells and THP-1.GILT cells infected with wild type *L. monocytogenes* or ΔLLO *L. monocytogenes*. Both strains of bacteria express GFP and are green. Phalloidin staining of actin is in red. **b**, Quantitation of actin polymerization. 1000 cells were counted at each time point and the percentage of infected cells with actin polymerization is indicated. The figure shows a representative result of two individual experiments.

Statistically significant time points are noted with * representing a $p<0.05$ and ** representing a $p<0.01$.

Supplemental Figure 4. Characterization of purified LLO. **a**, Purified LLO separated by reducing SDS-PAGE and stained with Coomassie Blue. **b**, Western blot of purified LLO separated by gradient SDS-PAGE using an LLO-specific rabbit antibody. The bands correspond to the expected Mr of a monomer, 58 kDa.

Supplemental Figure 5. Hemolytic activity of purified wild type SLO and a mutant SLO (C530A) lacking the single cysteine residue. a. Hemolytic activity of 300ng wild type SLO incubated with GILT or with GILT inactivated with NEM. The method was as described for LLO in Fig. 3d. **Average values from three individual experiments are shown and P values for significant differences indicated.** b. Hemolytic activities of wild type SLO (open bars) and the C530A SLO mutant (black bars). 300ng of each purified hemolysin was used with varying concentrations of GILT or with 4mM DTT (gray bars). **Average values from three individual experiments are shown and P values indicating significant different samples are indicated.**