SUPPLEMENTAL INFORMATION

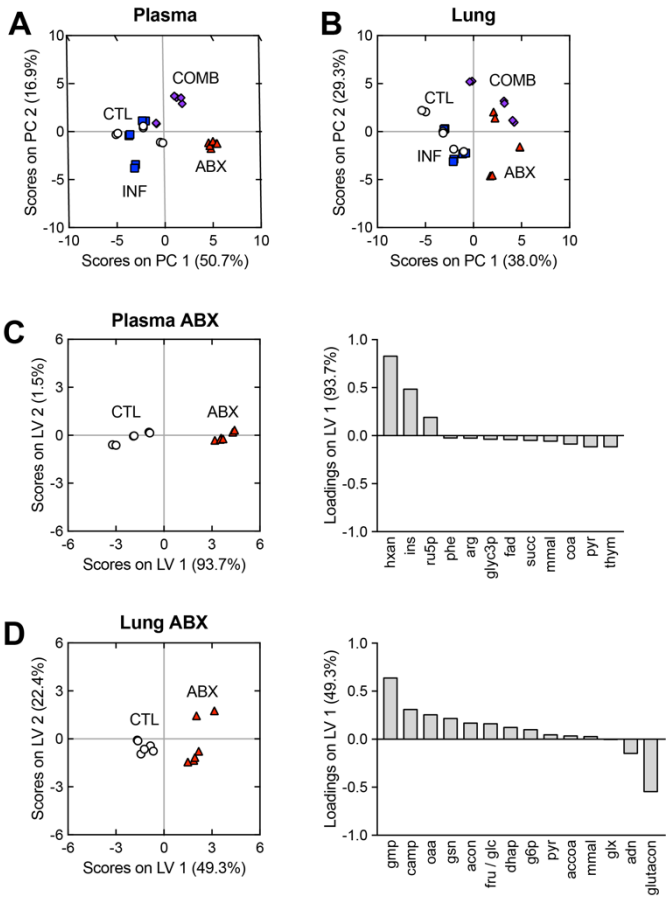

Figure S1. Antibiotic treatment systemically alters host metabolites; related to Figure 1.

(A) PCA projection of metabolomic profiles from plasma samples of all 4 treatment groups: control (CTL), antibiotic treated (ABX), infection (INF) or combination treatment (COMB).

(B) PCA projection of metabolomic profiles from lung samples of all 4 treatment groups.

(C) PLS-DA of plasma samples from ABX mice.

(D) PLS-DA of lung samples from ABX mice. Metabolites selected by elastic net regularization were enriched for carbohydrate metabolism intermediates.

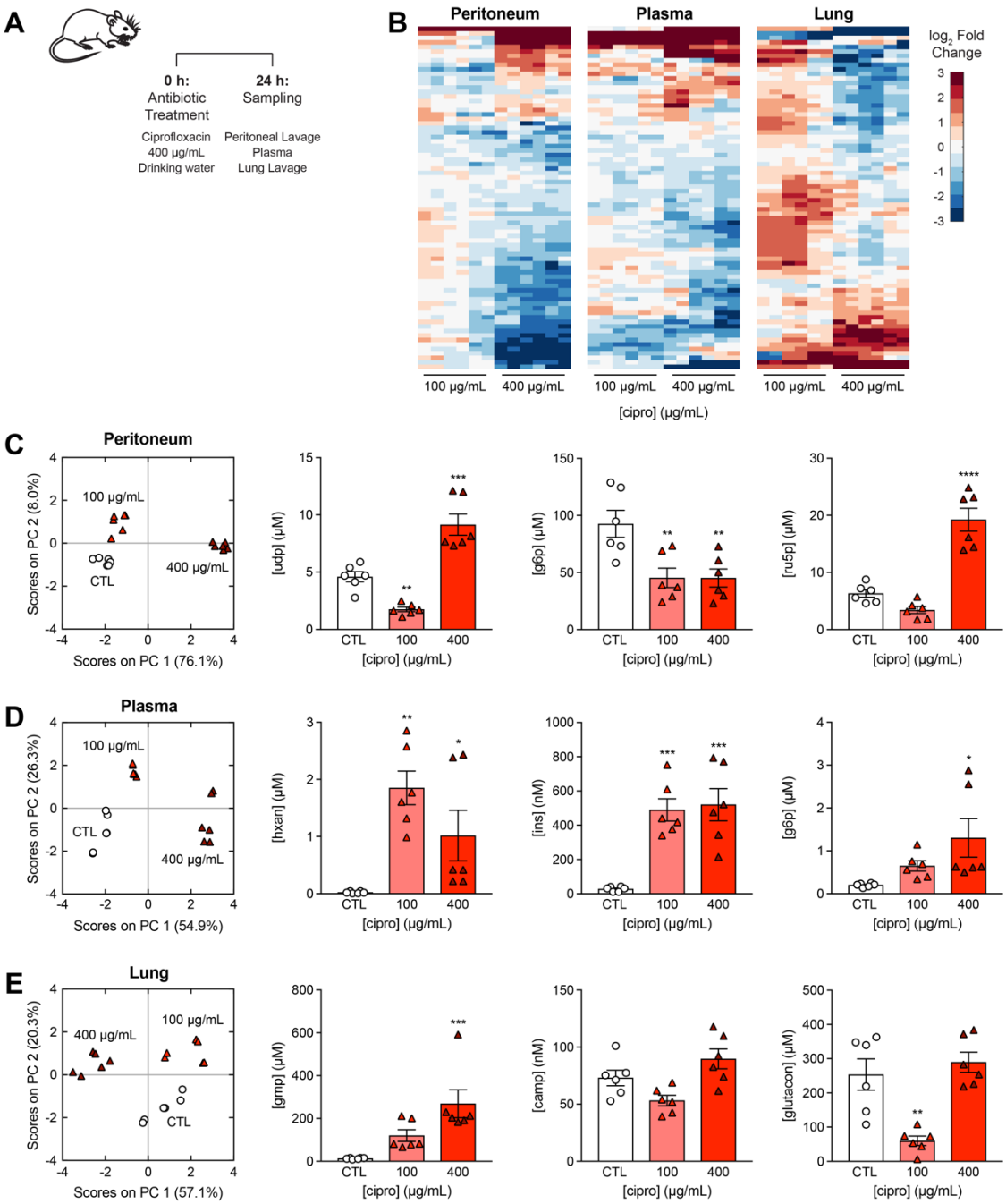

Figure S2. Antibiotic treatment elicits dose-dependent changes to host metabolites; related to Figure 1.

(A) Experimental design for metabolomic profiling. Conventional mice were subjected to antibiotic treatment with 400 µg/mL cipro and sampled 24 h later.

(B) Hierarchically clustered heatmaps for changes in metabolite concentrations between antibiotic treated and control mice at 100 and 400 µg/mL cipro, by tissue sampled.

(C) PCA projection of metabolomic profiles from control (CTL) and antibiotic treated mice in the peritoneum.

(D) PCA projection of metabolomic profiles from control (CTL) and antibiotic treated mice in the plasma.

(E) PCA projection of metabolomic profiles from control (CTL) and antibiotic treated mice in the lung.

Data are represented as mean ± SEM from n = 3 independent biological replicates. Significance reported as FDR-corrected p-values in comparison with corresponding CTL conditions: *: $p \leq 0.05$, **: $p \leq 0.01$, ***: $p \leq 0.001$, ****: $p \leq 0.0001$.

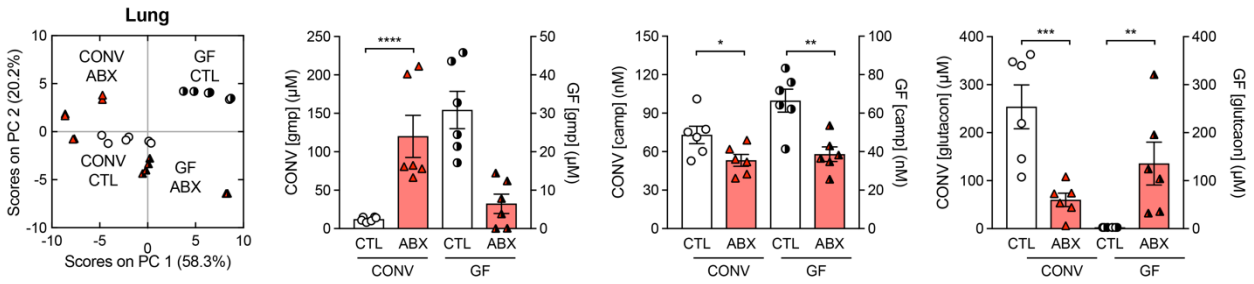

Figure S3. Antibiotic treatment elicits microbiome-independent changes in host metabolites; related to Figure 2. PCA projection of metabolomic profiles from control (CTL) and antibiotic treated (ABX) conventional (CONV) and germ-free (GF) mice in the lung (*left*). Concentrations for lung metabolites with large lung ABX LV1-loadings in CONV and GF mice (*right*). Data are represented as mean ± SEM from n = 3 independent biological replicates. Significance reported as FDR-corrected p-values in comparison with corresponding CTL conditions: *: $p \leq 0.05$, **: $p \leq 0.01$, ***: $p \leq 0.001$, ****: $p \leq 0.0001$.

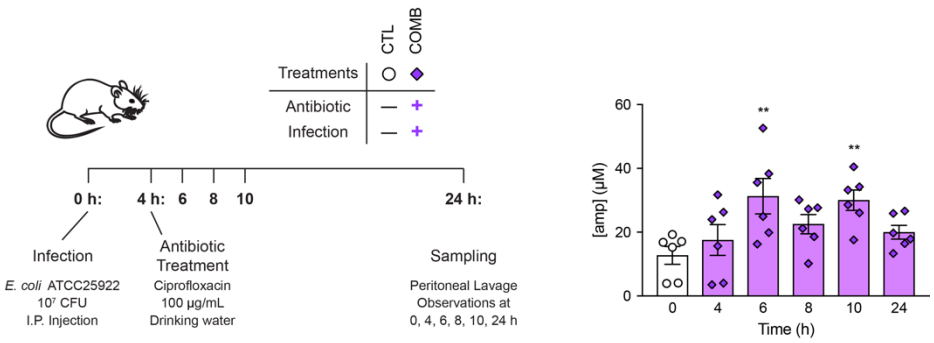

Figure S4. Metabolites altered by antibiotic treatment during infection inhibit drug efficacy; related to Figure 3. Peritoneal amp concentrations following antibiotic treatment. Conventional mice were subjected to antibiotic treatment with 100 µg/mL cipro and sampled at 4, 6, 8, 10 and 24 h. Data are represented as mean ± SEM from n = 3 independent biological replicates. Significance reported as p-values in comparison with corresponding CTL conditions: **: $p \leq 0.01$.

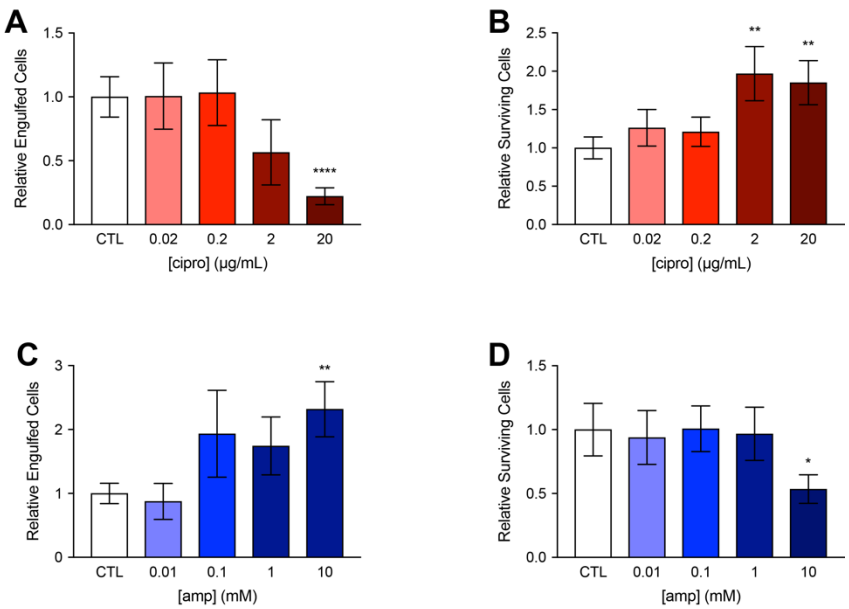

Figure S5. Antibiotic treatment directly inhibits phagocytic killing by immune cells; related to Figure 5.
(A) Pathogen engulfment by control (CTL) or macrophages treated with 0.2-20 µg/mL cipro.
(B) Pathogen survival by control or macrophages treated with 0.2-20 µg/mL cipro.
(C) Pathogen engulfment by control or macrophages treated with 0.01-10 mM amp.
(D) Pathogen survival by control or macrophages treated with 0.01-10 mM amp.
Data are represented as mean ± SEM from n ≥ 3 independent biological replicates. Significance reported as FDR-corrected p-values within the indicated comparisons: *: $p \leq 0.05$, **: $p \leq 0.01$, ****: $p \leq 0.0001$.