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Supplemental Material to:

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Cytosolic clearance of replication-deficient mutants reveals *Francisella tularensis* interactions with the autophagic pathway

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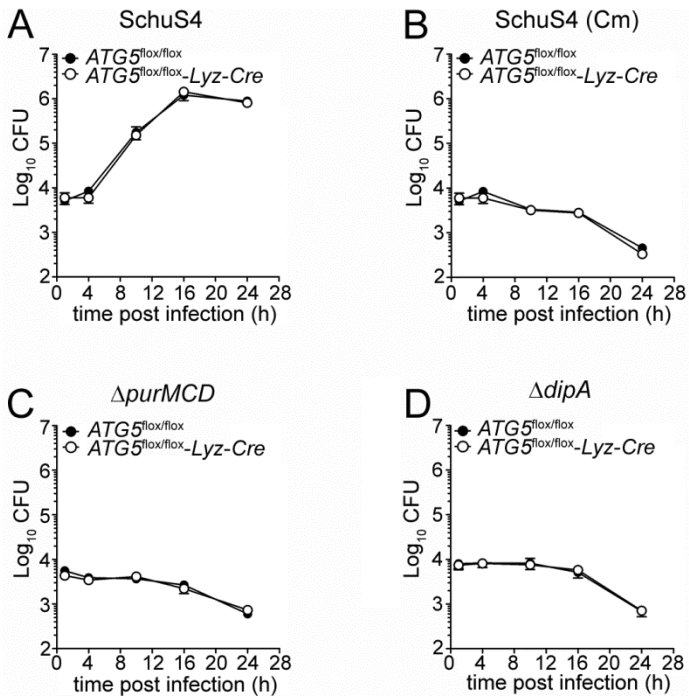

Figure S1. Intracellular growth of SchuS4 (A), SchuS4 (B) treated with chloramphenicol at 6 h pi, and its isogenic Δ*purMCD* (C) and Δ*dipA* (D) mutants in *ATG5*$^{flox/flox}$ and *ATG5*$^{flox/flox}$-Lyz-*Cre* BMMs. Intracellular bacteria were enumerated from CFUs at various times p.i. Data are means ± SD from a representative experiment performed in triplicate out of two independent repeats.

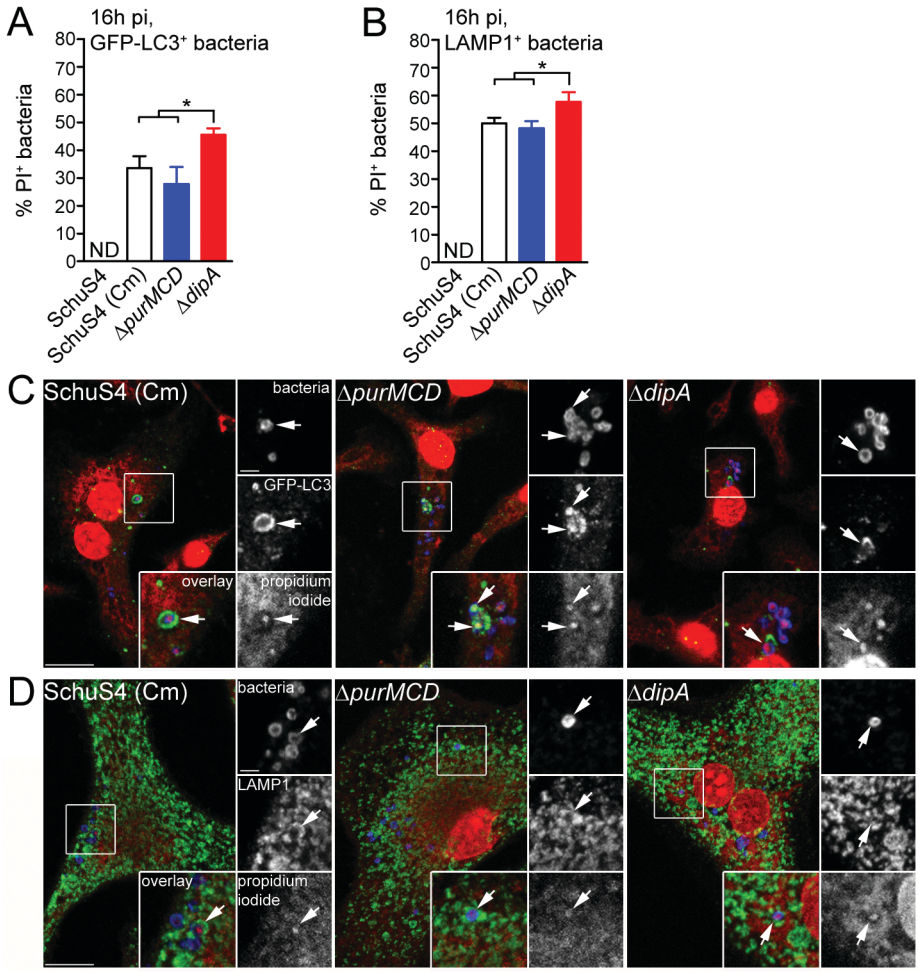

Figure S2. Viability of replication-deficient *Francisella* contained within autophagic vacuoles. (A and B) Quantification of LC3-positive (A) or LAMP1-positive (B) bacteria for propidium iodide labeling at 16 h p.i. Infected BMMs were subjected to an intracellular viability assay followed by immunofluorescence labelling for GFP (A) or LAMP1 (B). GFP-LC3- or LAMP1-positive bacteria were scored for propidium iodide labeling. At least 30 bacteria per experiment were scored for each condition. Data are means ± SD from three independent experiments. ND, not determined. Asterisks indicate statistically significant differences (*$p < 0.05$, two-tailed unpaired Student's t-test). (C and D) Representative confocal images of BMMs infected with either SchuS4 or its derivatives and processed for an intracellular viability assay (red) followed by immunofluorescence labeling of bacteria (blue), GFP (C, green) or LAMP1 (D, green) at 16 h p.i. Magnified insets show single channel images of the boxed areas. White arrows indicate propidium iodide-labelled bacteria enclosed within GFP-LC3- or LAMP1-positive vacuoles. Scale bars, 10 or 2 μm.