## Supplemental materials and methods

### Isovitalex Recipe

| Chemical                  | Stock concentration | Solvent          | Volume needed for 1 L |
|---------------------------|---------------------|------------------|-----------------------|
| Dextrose                  | 100 g/250 ml        | Water            | 250 ml                |
| Guanine HCl               | 30 mg/10 ml         | 0.1N NaOH        | 10 ml                 |
| L-glutamine               | 10g/250 ml          | Warm water       | 250 ml                |
| Thiamine HCl              | 9mg/30ml            | Water            | 10 ml                 |
| Adenine                   | 1g/50ml             | 0.2N HCl         | 50 ml                 |
| NAD                       | 250mg/10ml          | Water            | 10 ml                 |
| Vitamin B12               | 10mg/10ml           | Water            | 10 ml                 |
| 4-aminobenzoic acid (PABA)| 13mg/10ml           | Water            | 10 ml                 |
| Thiamine pyrophosphate    | 100mg/10ml          | Water            | 10 ml                 |
| Ferric nitrate            | 20mg/10ml           | Water            | 10 ml                 |
| L-cysteine                | 1.1g/100ml          | 0.5N HCl         | 100 ml                |
| L-cysteine HCl            | 25.9g/100ml         | Water            | 100 ml                |
Figure S1: (A) Growth kinetics of WT *F. tularensis* Schu S4 in CDM with DMSO or various doses of atglistatin. Data points are mean of 3 independent experiments performed in triplicate. (B) Representative confocal micrographs of WT MEFs treated overnight with 5 µM triacsin C, 10 µM T863, or 30 uM atglistatin. Cells were stained with BODIPY 493/503. Scale bar represents 5 µM. (C) Quantification of number of lipid droplets per cell (N=50 cells). Asterisk represents the significant difference as determined by one-way ANOVA and Dunnett's post-hoc test. *** p<0.001.
**Figure S2:** (A) Growth kinetics of WT *F. tularensis* Schu S4 harboring a luciferase plasmid (LUX) in J774A.1 macrophages. Luminescence (RLUs) was measured over 24 hrs. Data points are the mean from 3 independent biological replicates performed in triplicate. Asterisks represent significant difference between DMSO treated cells and 5 µM Compound C treated cells (blue) or 10 µM Compound C treated cells (purple) determined by one-way ANOVA and Dunnett’s post-hoc test. * p<0.05, ** p=0.004, *** p=0.002 (B) Growth kinetics of WT *F. tularensis* Schu S4 in CDM with DMSO or various doses of Compound C. Data points are mean of 3 independent experiments performed in triplicate. (C) Cell viability of *F. tularensis* infected J774A.1 with various doses of Compound C at 8 hrs. Data points are mean ± SD of 3 independent experiments performed in triplicate. Asterisks represent significant difference between DMSO control and Compound C treated cells determined by one-way ANOVA and Dunnett’s post-hoc test. (D) Cell viability of *F. tularensis* infected J774A.1 with various doses of Compound
C after 16 hrs of treatment. Data points are mean ± SD of 3 independent experiments performed in triplicate. Asterisks represent significant difference between DMSO control and Compound C treated cells determined by one-way ANOVA and Dunnett’s post-hoc test. Live and dead represent untreated cells. * p <0.05
**Figure S3:** (A) Representative confocal fluorescence micrographs of WT and AMPK -/- MEFs infected with GFP expressing WT *F. tularensis* Schu S4 or GFP expressing ΔdotU Schu S4 for 6 hrs pi. Nuclei were stained using DAPI. Scale bar represents 5 µm. (B) WT and AMPK -/- MEFs were infected as in (A). At 6hrs pi cells were fixed and immunostained for lysosomal marker LAMP-1. The number of bacteria associated with LAMP-1 staining was quantified by fluorescence microscopy. Data represents the mean ± SD of >3 independent experiments. N refers to the number of cells analyzed.
Figure S4: BMDMs infected with WT *F. tularensis* LVS S4 pEDL17 empty vector (EV), Δ*ripA* pEDL17 EV, and Δ*ripA::pEDL17 ripA*. Bacterial growth was measured via dilution plating at 4 hours and 24 hours pi. Cells were treated with 100 µg/ml of ATc at 12 hours pi to confirm plasmid and rescue growth of Δ*ripA::pEDL17 ripA*. Data points are the mean ± SD of 3 independent experiments performed in triplicate.