SUPPORTING INFORMATION

Figure S1. The effects of T3SS inhibitor on the development of *C. trachomatis* are significantly reduced in cells supplemented with exogenous iron. 0 or 250 μM $FeSO_4$ was added to HeLa cells at the time of infection with *Chlamydia trachomatis.* At 2 hpi or 8 hpi, DPP or C1 (respectively) was added. EBs were harvested at 44 hpi. Inclusion forming units (IFU) were then calculated by conducting serial dilutions, infecting fresh monolayers, and counting new inclusions at 24 hpi. The dotted line represents the limit of detection for the assay. Data plotted represent three independent biological replicates and error bars represent the standard deviations from the mean. **, $p < 0.01$.

Figure S2. The presence of immunostimulatory peptidoglycan is dependent on the timing of persistence induction. HEK hNOD1 SEAP-reporter assay comparing the abundance of shed, immunostimulatory peptidoglycan when iron sequestration is initiated at different time points throughout the *Chlamydia* developmental cycle. NOD activity was tested from cell supernatants at 24 hpi **(a)** and 36 hpi **(b)**. OD_{650} of the treatment group was normalized to the untreated and represented as percent SEAP activity. Data presented are the mean of three independent biological replicates and error bars represent standard deviation from the mean. **, $p < 0.005$; ns, not significant.

Figure S3: *Chlamydia* ABs differ in their presentation of early-stage inclusion membrane proteins. Immunolabeling of inclusion membrane proteins CT229 **(a)** and IncG **(b)** in *Chlamydia*-infected cells at 24 hpi. Inc presentation was compared for six different aberrance-inducing conditions. Cells treated with the protein synthesis inhibitor azithromycin at 8 hpi were used as a control. Images are representative of between 10-20 inclusions observed per condition over the course of three separate experiments. Scale bars; ~ 2 μm.

Figure S4: *Chlamydia* ABs differ in presentation of mid- and late- stage inclusion membrane proteins. Immunolabeling of inclusion membrane proteins IncA **(a)**, and CT813 **(b)** in *Chlamydia*-infected cells at 24 hpi. Inc presentation was compared for six different aberrance-inducing conditions with the protein synthesis inhibitor azithromycin used as a control. Scale bars; ~ 2 μm. Images are representative of between 10-20 inclusions observed per condition and the experiment was carried out three times.

Figure S5. Assessment of inclusion trafficking to the nucleus and actin cage formation. (a) The proximity of normal and aberrant *Chlamydia* inclusions to host cell nuclei was assessed by immunolabeling of normal and aberrant forms of *Chlamydia*. Nuclei were stained with Hoechst solution (blue) at 24 hpi. **(b)** The presence of actin cages about the inclusion periphery of normal and ABs of *Chlamydia* was assessed by staining cells with conjugated phalloidin at 46 hpi. *Chlamydia* was visualized utilizing MOMP monoclonal antibodies. Images are representative of 10-20 viewing planes carried out over three separate experiments. Scale bars; ~ 5 μm.

Graphical Abstract. Persistence halts the Chlamydia developmental cycle while simultaneously altering bacteria - host cell interactions. In this study, we provide evidence that non-dividing aberrant forms of *Chlamydia* differ in how they interact with host cells and our immune systems. The induction of persistence in response to a cell's innate response to infection results in *C. trachomatis* significantly reducing the synthesis and shedding of immunostimulatory peptidoglycan while concurrently minimizing the deleterious reorganization of host cell processes. We propose that this allows *C. trachomatis* to remain intracellular and escape clearance by the cell-mediated immune response for extended periods of time while simultaneously enhancing the longevity of an infected host cell.