

### Investigations into coinfections of the obligate intracellular ruminant pathogens *Chlamydia abortus* and *Coxiella burnetii*

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*Chlamydia abortus* and *Coxiella burnetii* are obligate intracellular gram negative bacteria that infect small ruminants. Both target the placenta, can cause abortion and possess a zoonotic potential. Chlamydia and Coxiella share even more striking similarities on the cellular and molecular level such as a biphasic life-cycle with extracellular, infectious variants and intracellular, non-infectious forms residing in a membrane-bound vacuole. By hijacking intracellular organelles and redirecting transport vesicles, the bacteria acquire essential nutrients, but in a different mode. While the vacuole of Coxiella is an acidified phagolysosome that fuses with endocytic vesicles, Chlamydia in non-acidified inclusions receives its nutrients from fusiogenic events with exocytic vesicles from the Golgi apparatus and the endoplasmic reticulum. Field studies in small ruminants have shown coinfections of Chlamydia and Coxiella in placental tissue from abortions. We have screened 65 placenta samples collected after normal parturition from infected sheep flocks. 52.3 % of these samples were PCR-positive for *Chl. abortus*, 61.5 % for *Cox. burnetii* and in 40.0 %, a coinfection of both agents was detected. To investigate whether the interaction of the two pathogens is of synergistic, competitive or neutral nature and to better assess the contribution of such polymicrobial infections to disease progression, we analyzed the interaction of *Chl. abortus* DC59 and *Cox. burnetii* RSA 439 NMII in cell culture models. Fluorescence and electron microscopy revealed that different cell lines can be coinfecting with Coxiella and Chlamydia and that a single cell can harbor both pathogens. They reside in distinct vacuoles but in close proximity to each other with occasional fusion of vacuole membranes. A preinfection of cells with Coxiella does not alter general Chlamydia morphology, but growth and infectivity was negatively influenced as shown by qPCR analysis of DNA replication and titration.

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