

## Supplemental Figure Legend

**S1.** Vero cells infected by *Rickettsia monteiroi* as demonstrated by Gimenez staining (A and B), by immunofluorescence assay using guinea pig anti- *Rickettsia monteiroi* serum (1:64 dilution) and anti-guinea pig immunoglobulin G secondary antibody (C), and by immunofluorescence assay using guinea pig anti-*Rickettsia bellii* serum (1:64 dilution) and anti-guinea pig immunoglobulin G secondary antibody (D). Photographs were taken using an Olympus optical microscope with a 10× ocular and a 100× objective (A and B) and an Olympus fluorescence microscope with a 10× ocular and a 40× objective (C and D).

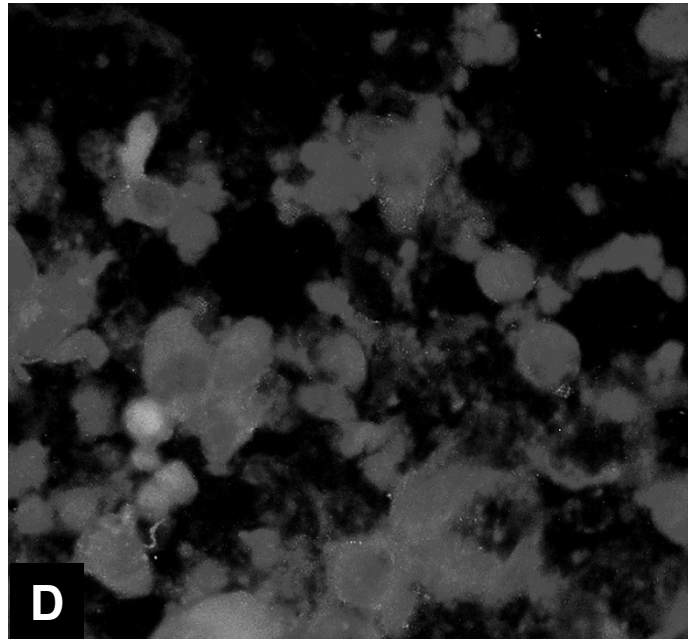
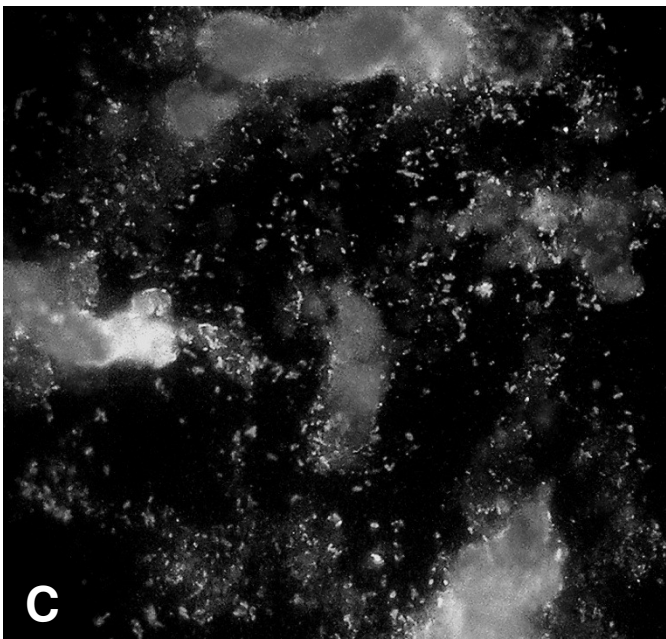
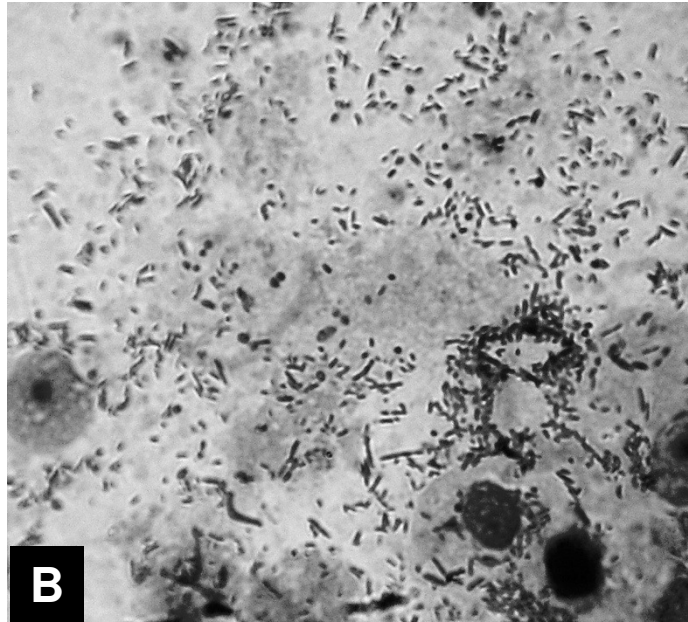
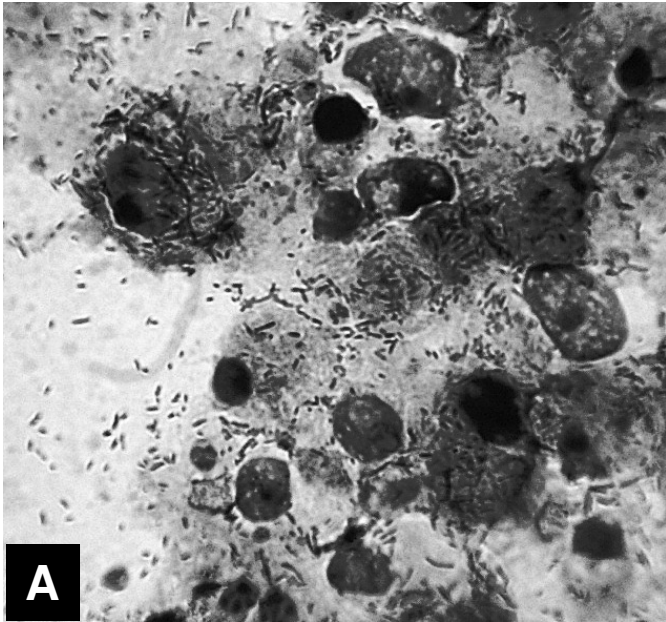
**S2.** Molecular phylogenetic analysis of *Rickettsia monteiroi* isolated from the tick *Amblyomma incisum* from Brazil. A total of 1,321 unambiguously aligned nucleotide sites of the rickettsial gene *rrs* were subjected to analysis. Bootstrap values higher than 50% are shown at the nodes. Numbers in brackets are GenBank accession numbers. Bar, 0.1% difference.

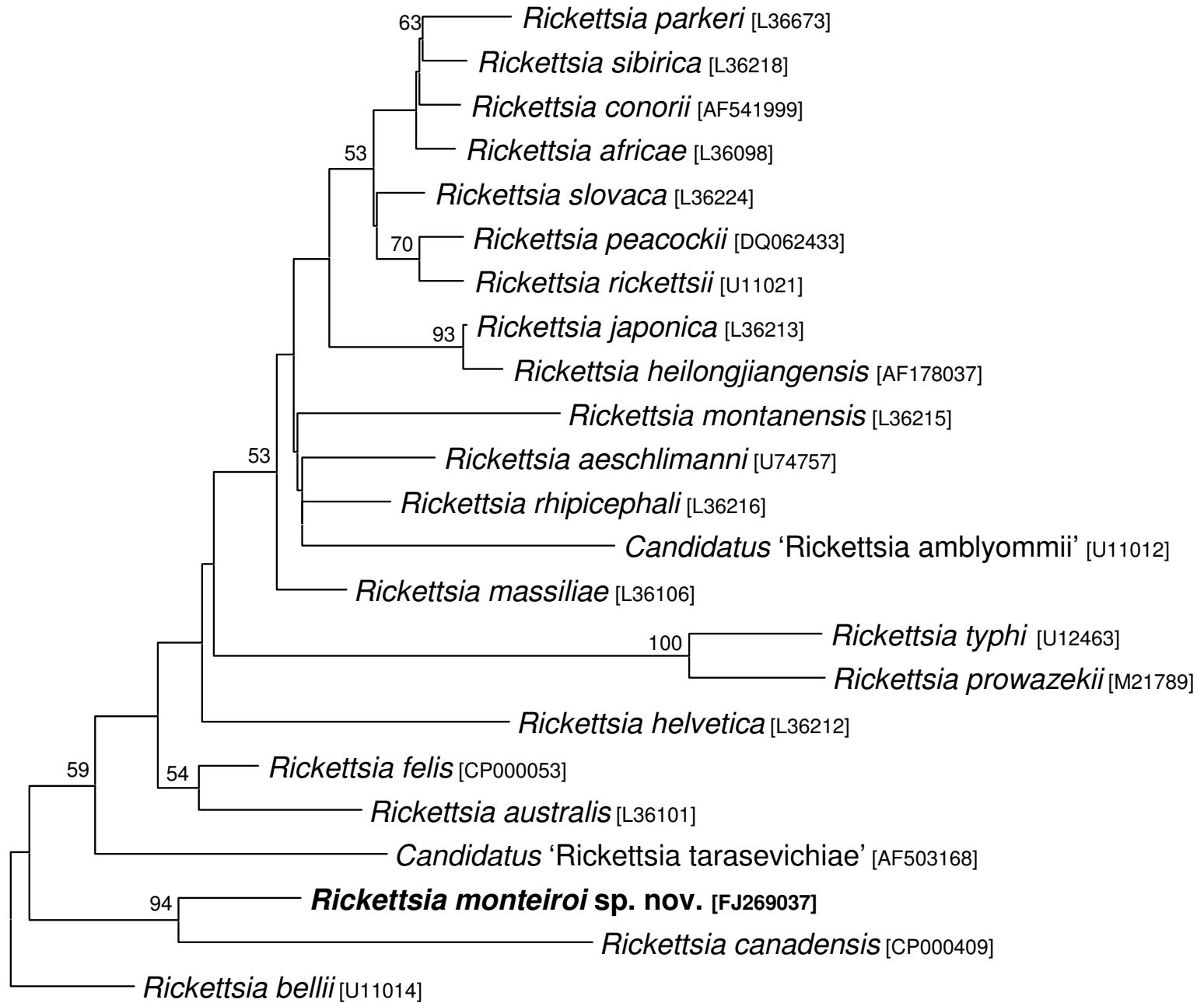
**S3.** Molecular phylogenetic analysis of *Rickettsia monteiroi* isolated from the tick *Amblyomma incisum* from Brazil. A total of 829 unambiguously aligned nucleotide sites of the rickettsial gene *gltA* were subjected to analysis. Bootstrap values higher than 50% are shown at the nodes. Numbers in brackets are GenBank accession numbers. SFG: spotted fever group; TRG: transitional group; TG: typhus group; CG: canadensis group; AG: adalia group; BG: bellii group. Bar, 2% difference.

**S4.** Molecular phylogenetic analysis of *Rickettsia monteiroi* isolated from the tick *Amblyomma incisum* from Brazil. A total of 386 unambiguously aligned nucleotide sites of the rickettsial gene *htrA* were subjected to analysis. Bootstrap values higher than 50% are shown at the nodes. Numbers in brackets are GenBank accession numbers. Bar, 1% difference.

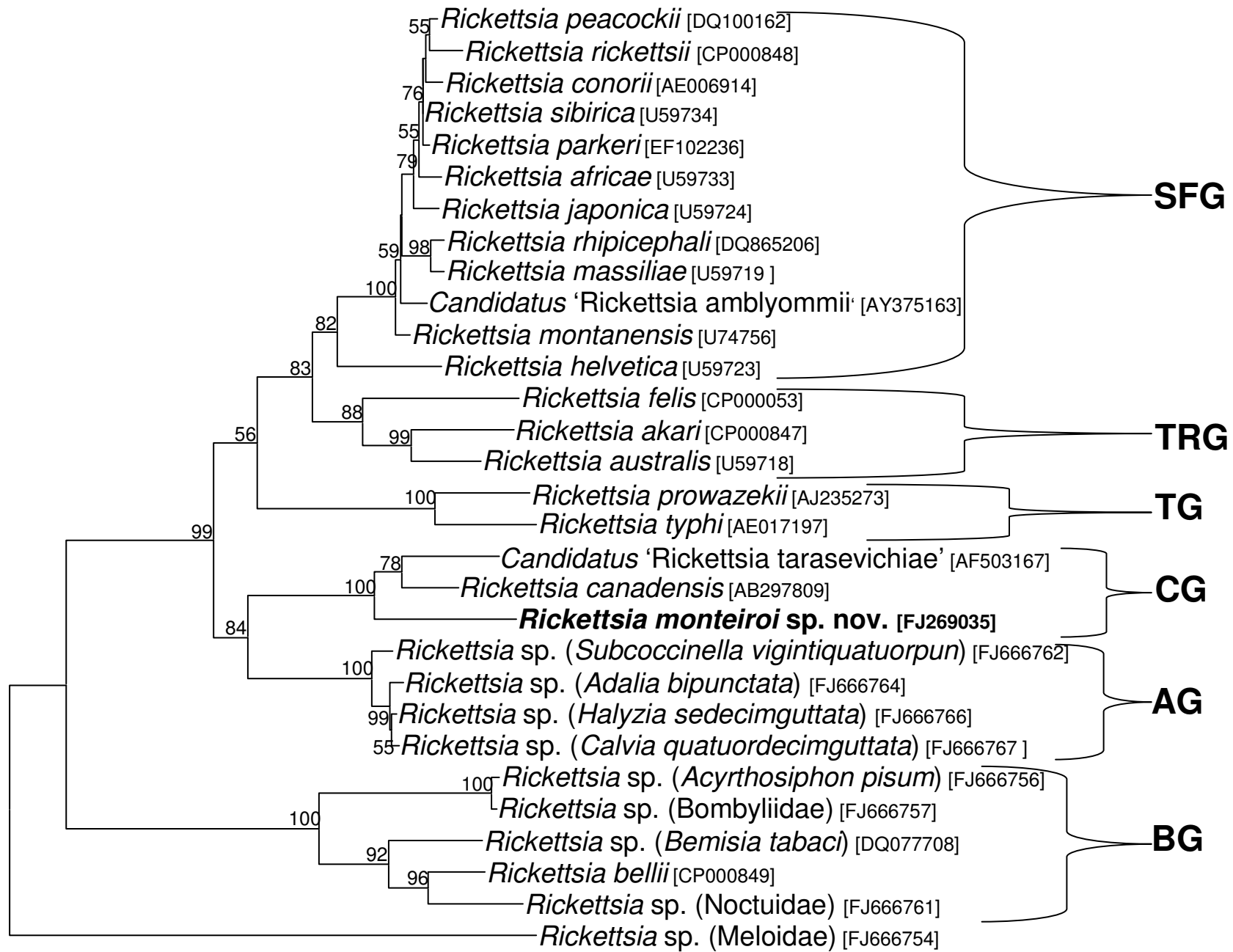
**S5.** Molecular phylogenetic analysis of *Rickettsia monteiroi* isolated from the tick *Amblyomma incisum* from Brazil. A total of 443 unambiguously aligned nucleotide sites of the rickettsial gene *sca1* were subjected to analysis. Bootstrap values higher than 50% are shown at the nodes. Numbers in brackets are GenBank accession numbers. Bar, 0.5% difference.

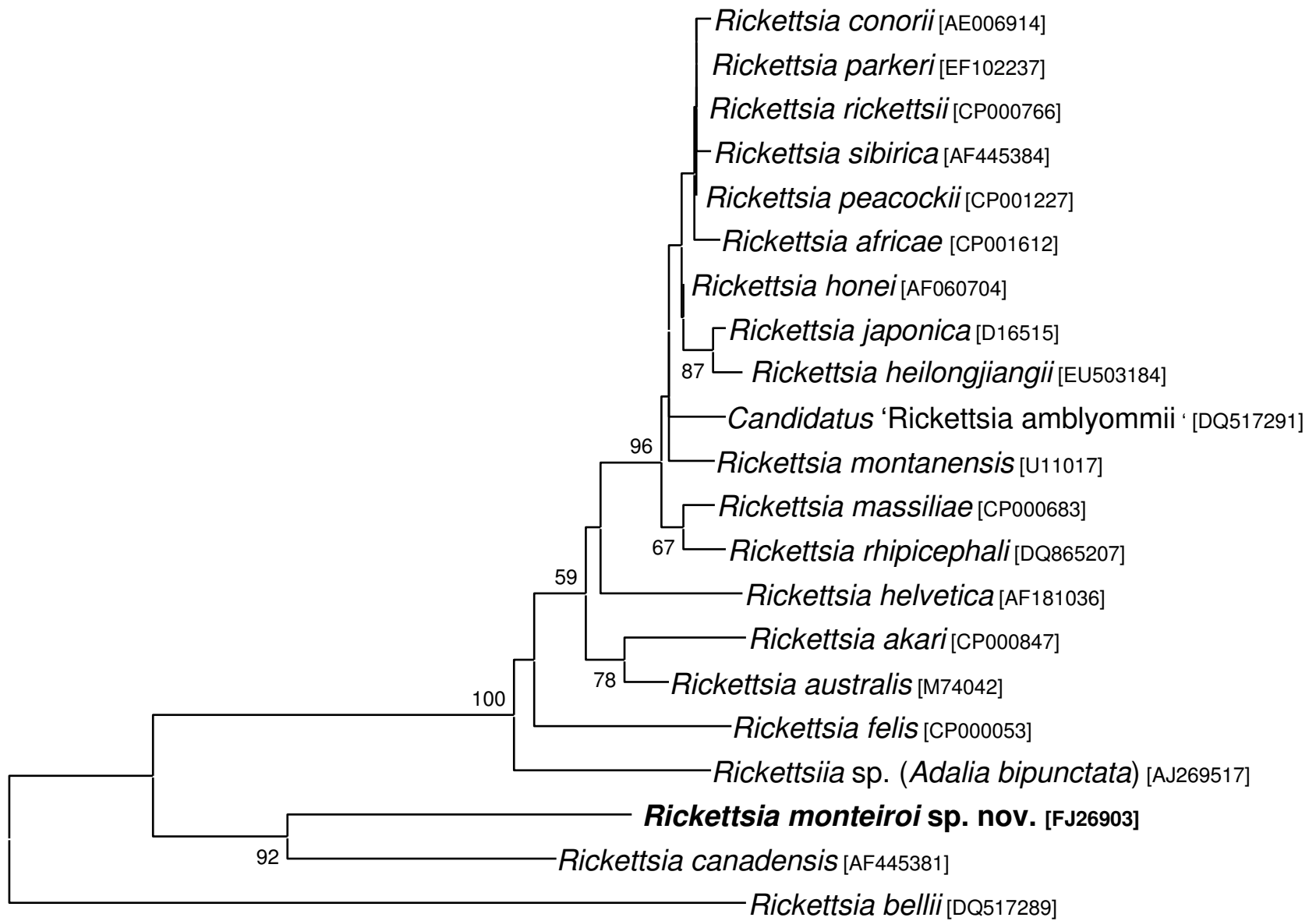
**S6.** Transmission electronic microscopy of *Rickettsia monteiroi* -infected Vero cells. (A) Several electron dense rickettsiae (black arrows) free in the cytosol of a Vero cell. N: nucleus; Bar, 2.5 µm. (B) A rickettsia showing characteristic Gram-negative morphology, with a cell wall (arrow head) and a cytoplasmic membrane (black arrow) separated from the cell wall by the periplasmic space. Bar, 0.34 µm.





0.1%





1%

