

Supplemental Material

Tab. S1 No cross-reactivity was observed with any of the tested bacteria, confirming specificity of the newly designed assays (ATCC: American Type Culture Collection; DSMZ: German Collection of Microorganisms and Cultures; NCTC: National Collection of Type cultures)

Species	Strain
<i>Acinetobacter baumanii</i>	DSMZ 7324
<i>Bacillus anthracis</i>	Volum
<i>Bacillus cereus</i>	ATCC 10987
<i>Bacillus globigii</i>	DSMZ 7264
<i>Bacillus thuringensis</i>	DSMZ 2046
<i>Brucella abortus</i>	ATCC 23448
<i>Burkholderia cepacia</i>	NCTC 10744
<i>Burkholderia mallei</i>	Zagreb
<i>Burkholderia pseudomallei</i>	ATCC 23343
<i>Burkholderia thailandensis</i>	DSMZ 13276
<i>Campylobacter jejuni</i>	ATCC 29482
<i>Candida albicans</i>	ATCC 36232
<i>Chlamydophila pneumoniae</i>	ATCC 53592
<i>Citrobacter freundii</i>	DSMZ 30039
<i>Clostridium perfringens</i>	ATCC 12915
<i>Clostridium sporogenes</i>	DSMZ 795
<i>Coxiella burnetii</i>	Nine Mile
<i>Eikenella corrodens</i>	DSMZ 8340
<i>Enterobacter aerogens</i>	DSMZ 30053
<i>Enterobacter faecalis</i>	DSMZ 2570
<i>Escherichia coli</i>	ATCC 11303

Species	Strain
<i>Haemophilus influenzae</i>	ATCC 10211
<i>Klebsiella pneumoniae</i>	ATCC 13883
<i>Legionella pneumophila</i>	NCTC 10332
<i>Listeria monocytogenes</i>	DSMZ 12464
<i>Moraxella catarrhalis</i>	DSMZ 9143
<i>Mycobacterium tuberculosis</i>	
<i>Neisseria meningitidis</i>	Patient isolate
<i>Propriionibacterium acnes</i>	DSMZ 1897
<i>Proteus mirabilis</i>	DSMZ 788
<i>Pseudomonas aeruginosa</i>	ATCC 10145
<i>Salmonella enterica typhii</i>	20-3267
<i>Serratia marcescens</i>	DSMZ 1636
<i>Shigella dysinteriae</i>	B476
<i>Staphylococcus aureus</i>	DSMZ 19041
<i>Staphylococcus epidermidis</i>	DSMZ 1798
<i>Stenotrophomoas maltophilia</i>	ATCC 5131
<i>Streptococcus pneumoniae</i>	DSMZ 20566
<i>Streptococcus pyogenes</i>	DSMZ 20565
<i>Vibrio cholerae</i>	ATCC 15748
<i>Yersinia enterocolitica</i>	DSMZ 13030
<i>Yersinia frederiksenii</i>	ATCC 33641
<i>Yersinia pseudotuberculosis</i>	ATCC 29833

Tab. S2 Overview of various *F. tularensis* and *Y. pestis* strains, which were tested for DNA detection capabilities of the newly designed assay via rtPCR. (ATCC: American Type Culture Collection; DSMZ: German Collection of Microorganisms and Cultures; IMB: Bacterial strain collection of the Bundeswehr Institute of Microbiology; NCTC: National Collection of Type cultures)

Species	Strain
<i>Francisella tularensis hispaniensis</i>	DSMZ 22475
<i>Francisella tularensis holartica japonica</i>	IMB FO700285
<i>Francisella noatunesis noatunesis</i>	DSMZ 23596
<i>Francisella philomiragia</i>	ATCC 25016
<i>Francisella tularensis mediasiatica</i>	FSC 149
<i>Francisella tularensis novicida</i>	FSC 595
<i>Francisella tularensis novicida</i>	FSC 156
<i>Yersinia pestis antiqua</i> 1.ANT	IMB L3-2996
<i>Yersinia pestis antiqua</i> 2. ANT	IMB L3-3004
<i>Yersinia pestis medievalis</i>	IMB L3-2536
<i>Yersinia pestis orientalis</i>	IMB L3-2540
<i>Yersinia pestis orientalis</i>	IMB L3-2992
<i>Yersinia pestis pestoides</i>	IMB L3-2997

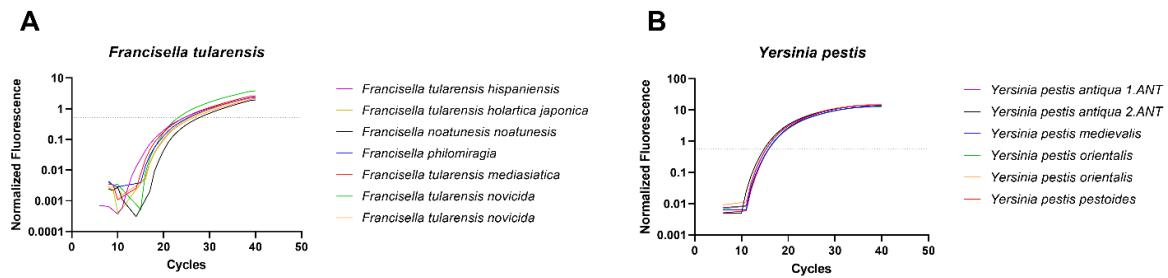


Figure S1 Real time PCR (rtPCR) detection of several (A) *Francisella tularensis* (*F. tularensis*) and (B) *Yersinia pestis* (*Y. pestis*) strains by using the newly designed assay, confirming the ability to detect in general *F.tularensis* and *Y.pestis* DNA.

The rtPCR was conducted as described in the part for Materials and Methods. 5 µl DNA was added to each rtPCR. For *Y. pestis* rtPCR, the competitive probe was included (*Y. pestis* probe:competitive probe 1:3). The first five cycles were excluded and the software calculated the threshold automatically. All measurements were performed in duplicates and repeated three times. The mean was estimated for each strain and is monitored for (A) *F. tularensis* and (B) *Y.pestis* strains.