

Electronic Supplementary Material

Multiplex isothermal solid-phase recombinase polymerase amplification for the specific and fast DNA-based detection of three bacterial pathogens

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On chip RPA experiments targeting a single pathogen:

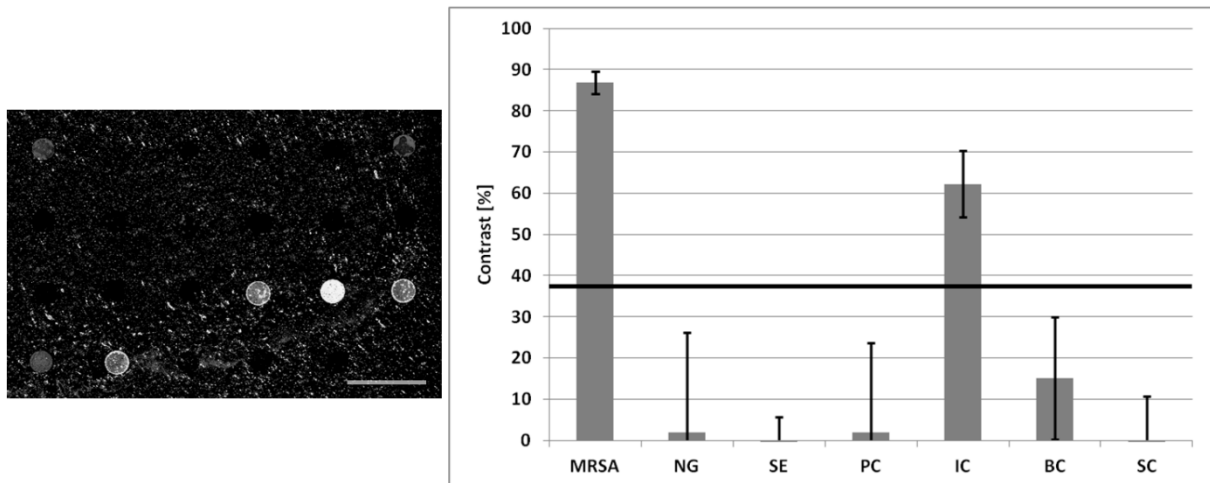


Fig. S1a On chip RPA experiment for the detection of methicillin-resistant *Staphylococcus aureus* (MRSA). Laser scanner images of one subarray (left side) and quantitative analysis (right side); Dynamic limit of detection (LOD) (horizontal line), IC: immobilization control; BC: buffer control; SC: specificity control.

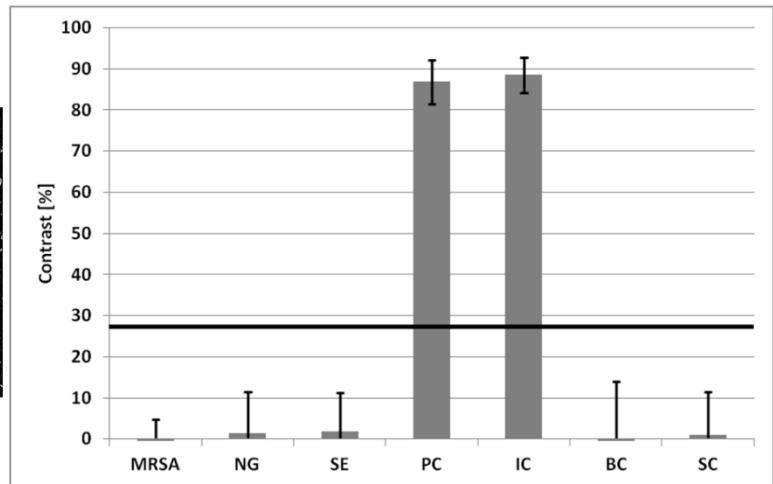
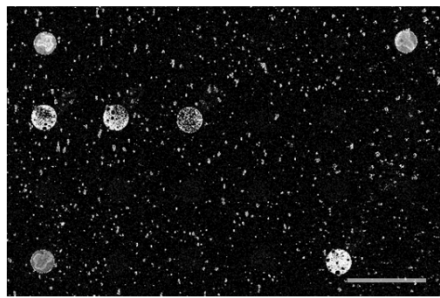


Fig. S1b On chip RPA experiment for the amplification of a plasmid control. Laser scanner images of one subarray (left side) and quantitative analysis (right side); Dynamic limit of detection (LOD) (horizontal line), IC: immobilization control; BC: buffer control; SC: specificity control.

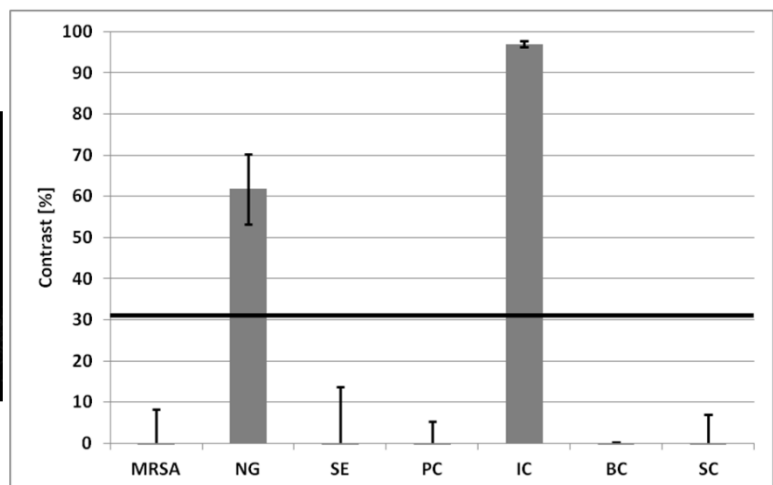
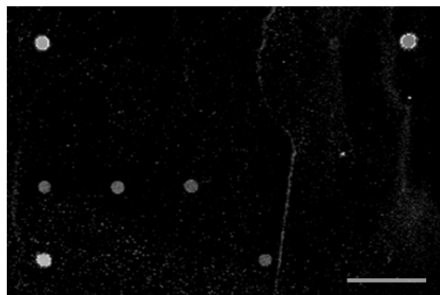


Fig. S1c On chip RPA experiment for the detection of *Neisseria gonorrhoeae* (NG). Laser scanner images of one subarray (left side) and quantitative analysis (right side); Dynamic limit of detection (LOD) (horizontal line), IC: immobilization control; BC: buffer control; SC: specificity control.

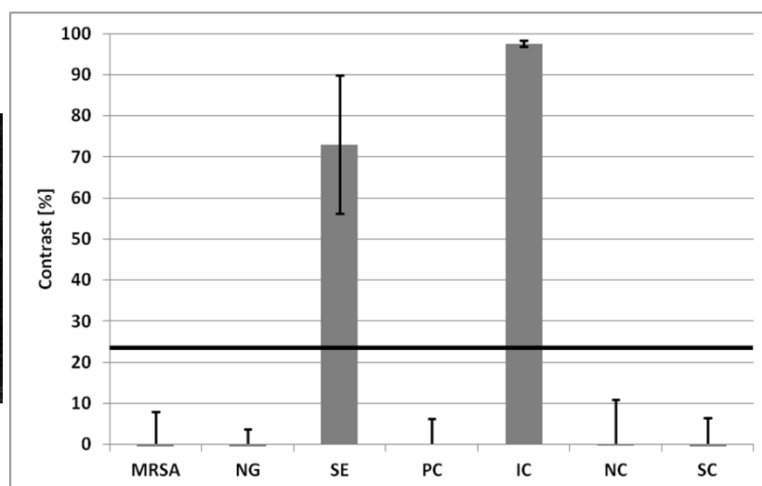
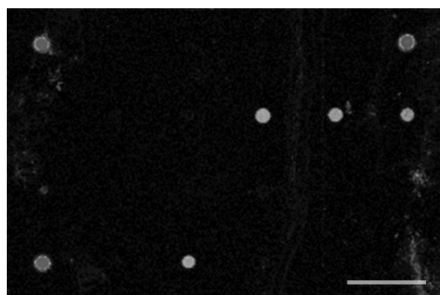


Fig. S1d On chip RPA experiment for the detection of *Salmonella enterica* (SE). Laser scanner images of one subarray (left side) and quantitative analysis (right side); Dynamic limit of detection (LOD) (horizontal line), IC: immobilization control; BC: buffer control; SC: specificity control.

Negative control for multiplex on chip RPA experiments:

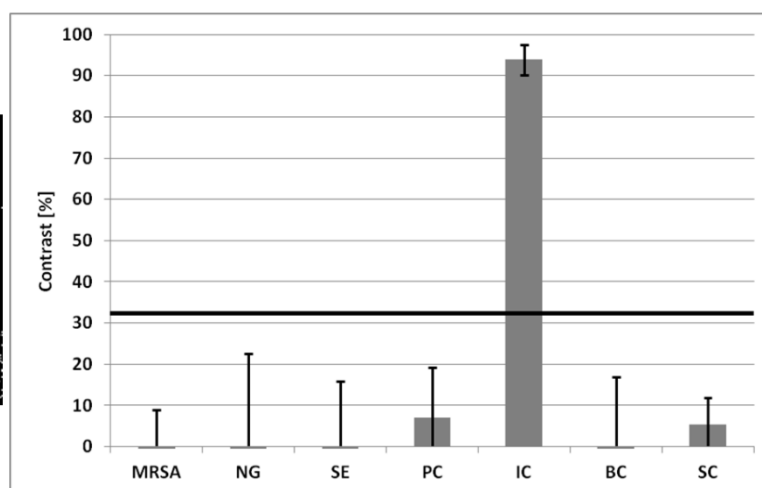
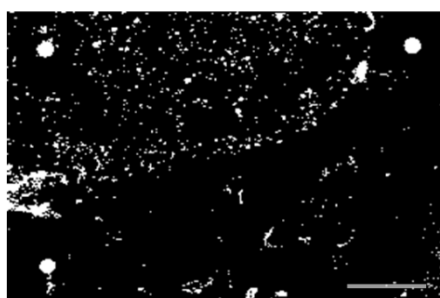


Fig. S1e Control quadruplex on chip RPA experiment using all primer pairs but not template DNAs. Laser scanner images of one subarray (left side) and quantitative analysis (right side); Dynamic limit of detection (LOD) (horizontal line), IC: immobilization control; BC: buffer control; SC: specificity control.

Sensitivity of the on chip RPA:

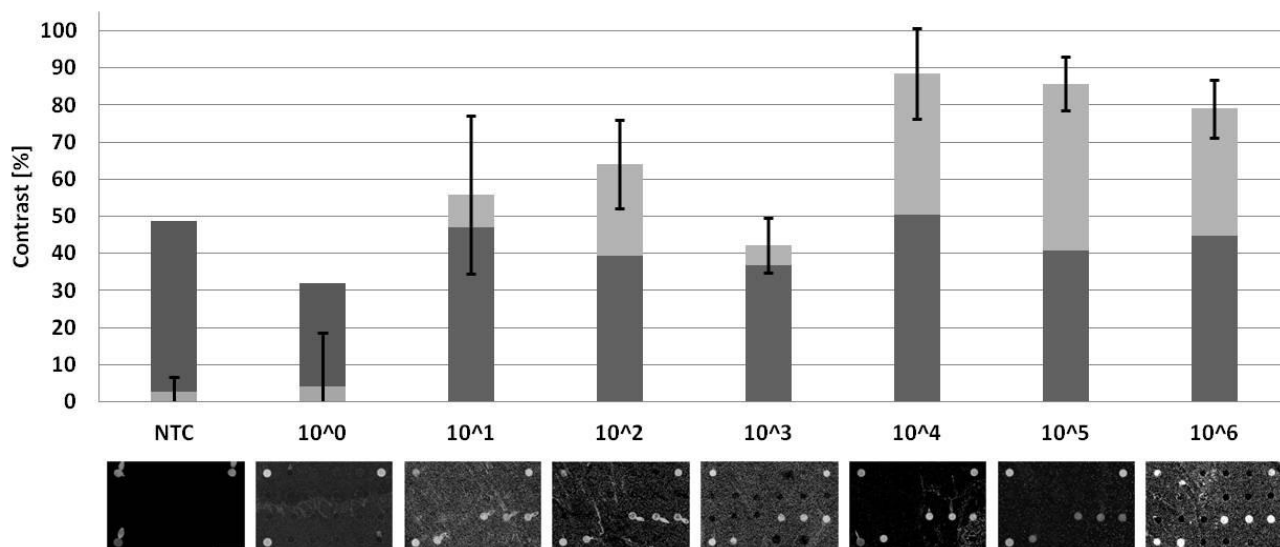


Fig. S2 Sensitivity test for the on chip RPA assay with a serial dilution of genomic DNA from methicillin-resistant *Staphylococcus aureus* from 1 to 10⁶ colony forming units (CFU) indicating a successful amplification with 10 copies. NTC: no template control; dark grey columns: dynamic LOD for every assay; light grey columns: Contrast values for MRSA specific signals on slides.

Inhibition experiments:

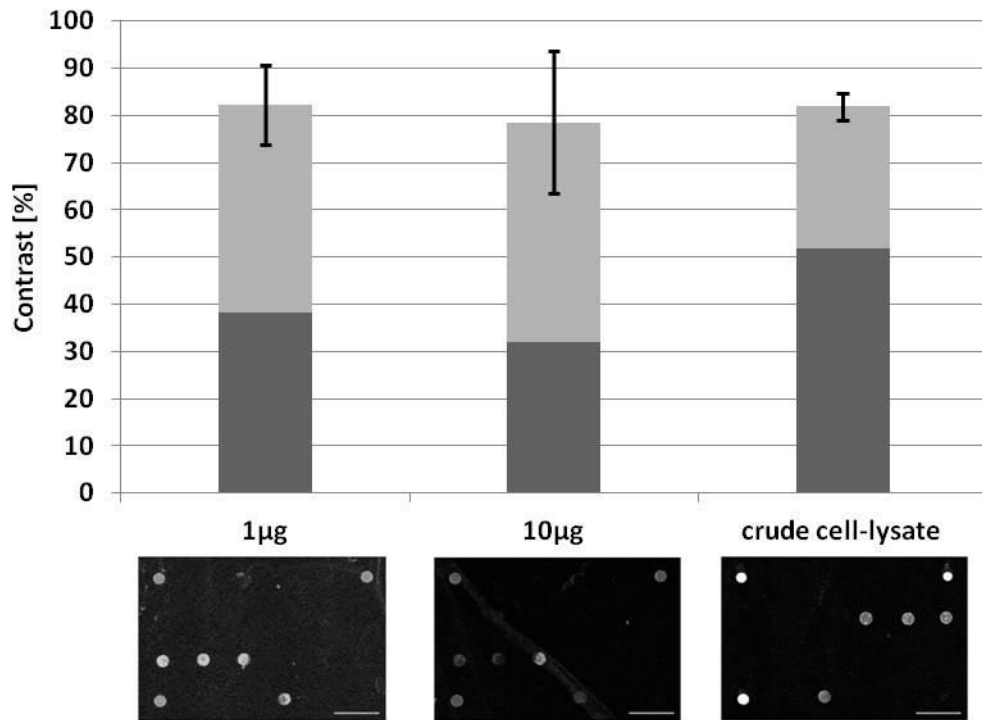


Fig. S3 Inhibition experiments with the on chip RPA show no significant reduction of signal when amplified in complex reaction conditions. 1 ng of gDNA of *Neisseria gonorrhoeae* was spiked into 1 µg and 10 µg of salmon sperm DNA and on chip RPA experiments were conducted according to the previously described procedure. 1 ng of gDNA of *Salmonella enterica* was spiked into a crude cell lysate of 2×10^4 Jurkat cells and used for on chip RPA without further treatment. Dark grey: Independently calculated dynamic limit of detection for every assay. Light grey: Signal for *Neisseria gonorrhoeae* or *Salmonella enterica*-specific positions on slides.