DNA-based detection of human pathogen water contaminants

<u>Cornelia Reuter</u>, Manuel Arnold, Nicole Slesiona, Matthias Urban, Andrea Csáki, Wolfgang Fritzsche

Leibniz Institute of Photonic Technology, Albert-Einstein-Str. 9, 07745 Jena, Germany

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Frequent outbreaks of water- and foodborne pathogens constitute a serious public health risk. One bacterial agent *Legionella pneumophila* (*L. pneumophila*) is the causative pathogen of Legionnaires' disease and can occur in almost every drinking water installation. Quick and precise detection results, in small scales, can be achieved with molecular methods. E.g. polymerase chain reaction (PCR) [1], loop mediated isothermal amplification (LAMP), recombinase polymerase amplification (RPA), in digital droplet applications, and fast colorimetric assays utilizing gold nanoparticles (AuNP).

In this work isothermal DNA amplifications were optimized, focusing on short reaction times, specificity, and promising high sensitivity, and studied for further investigations [2]. We were extending the established DNA assays towards a colorimetric assay, detecting genomic DNA of *L. pneumophila* using colloidal gold nanoparticles (Figure 1b). Non-target and negative DNA samples after amplification show a shift in absorption maxima simultaneous with a visible color change from red to blue in absence of target DNA, induced by coalescence (Figure 1c). This is measured with a cost efficient 2-LED- μ Spectrophotometer (Figure 1a), using two wavelengths and standard disposable UVVIS-cuvettes. The presented method is highly specific and point-of-care-compatible (PoC). Another approach is the generation of droplets in the picoliter scale. The number of positive and negative signals (with and without target), of L. *pneumophila*, is converted into copy numbers by applying Poisson statistics. This allows an absolute DNA quantification, independent from device settings and standard curves.

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Figure 1: (a) 2-LED-µSpectrophotometer using two wavelengths and standard UVVIS-cuvettes. (b) Selection of coloidal AuNP solution samples containing positive isothermal DNA amplification reaction (red), negative control (blue) and MgCl₂. (c) Corresponding absorbance measurements.

[1] C. Reuter et al., Lab on a Chip submitted (2019)

[2] C. Reuter et al., *Water Research* submitted (2019)