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Pauline Chevalier, Juliette Nourry, Delphine Aldebert, Eric Peyrin, Farid Oukacine, Basile Peres, Luc Choisnard

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Aptamer-based kit for the specific quantification of microorganisms

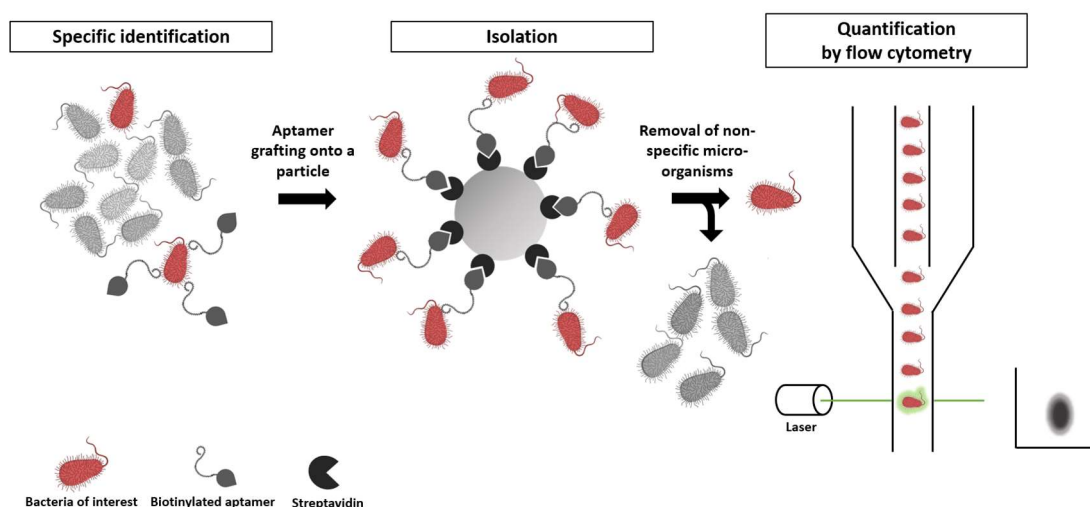
Authors: Pauline Chevalier & Juliette Nourry, Delphine Aldebert, Eric Peyrin, Farid Oukacine, Basile Peres, Luc Choisnard

Controlling the spread of bacterial infectious diseases is a major public health issue, notably because of the emergence of multi-resistant and emerging highly resistant bacteria. The Centre for Disease Control (CDC) reports that in the United States, more than 2,8 million individuals contract antibiotic-resistant bacteria annually, resulting in approximately 35,000 deaths [1]. Regarding that, detecting, identifying and quantifying microorganisms is a powerful prophylaxis, which plays a part in the fight against antibiotic resistance. However, current reference methods are time-consuming because they rely on the culture of the micro-organism. The turnaround-time is generally around 24 to 72 hours, which is highly incompatible with public health issues, especially if the diagnosis is of vital importance.

In that context, our aim is to propose a complementary detection tool to overcome the deficiencies of reference methods. The proposed method offers rapidity, specificity, sensitivity and versatility. The project is divided in three steps: the specific identification of the micro-organism of interest in mixture with others, the concentration and the extraction of the microorganism using an approach based on coupling the aptamer to a solid support that can be secondarily isolated from the contaminated sample, and then the quantification of the micro-organism by flow cytometry.

The aim of this communication is to present the development of the first step of the project. To implement the first step of identification, several aptamer sequences from the literature were used to detect *Escherichia coli* strain O157:H7, as used as model. The detection of the bacteria of interest by its specific aptamer is assessed by flow cytometry. Using a batch of aptamers labelled with a fluorophore, it is possible to discriminate bacteria capable of interacting via molecular patterns on their surface. A concentration range of aptamer from 0 to 400 nM was tested. It was achievable to obtain up to 60% of labelled bacteria. This clearly indicates recognition between the aptamer and the bacteria. The specificity of this signal was then determined by testing other strains than *E. coli*, such as *Acinetobacter baumannii* and *Streptococcus pneumoniae*.

Although a certain number of interactions between aptamers and their bacterial targets have been identified, as reported in the literature, specificity studies have so far failed to demonstrate the specificity of these interactions.



[1] CDC, Antibiotic resistance threats in the United States, 2019, Atlanta, Georgia, 2019. <https://doi.org/10.15620/cdc:82532>.