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Screening of specific aptamers for *Bacillus cereus* by Surface Plasmon Resonance imaging (SPRI)



Mathilde MANCEAU^{1,*}, Carole FARRE¹, Florence LAGARDE¹, Raphaël MATHEY², Arnaud BUHOT², Jasmina VIDIC³, Yanxia HOU-BROUTIN², Carole CHAIX^{1,*}

¹ Insitute of Analytical Sciences, UMR 5280 CNRS, University of Lyon, University Claude Bernard Lyon 1, 69100, Villeurbanne

² University Grenoble Alpes, CEA, CNRS, IRIG-SyMMES, 38300, Grenoble

³ INRAE, AgroParisTech, Insitute Micalis, University Paris-Saclay, 78350, Jouy-en-Josas

* mathilde.manceau@isa-lyon.fr ; carole.chaix-bauvais@univ-lyon1.fr

INTRODUCTION

Bacillus cereus is a **food poisoning** bacterium present in dairy products. To detect it, antibody based biosensors are efficient but available antibodies are expensive and may cross-react with other *Bacillus* species. **Aptamers** are a good alternative to these issues. Herein, seven aptamer sequences derived from the literature^(1,2) are **compared** and their specificity to *B. cereus* is assessed versus *Escherichia coli* and *Bacillus subtilis* by **Surface Plasmon Resonance imaging (SPRI)**.

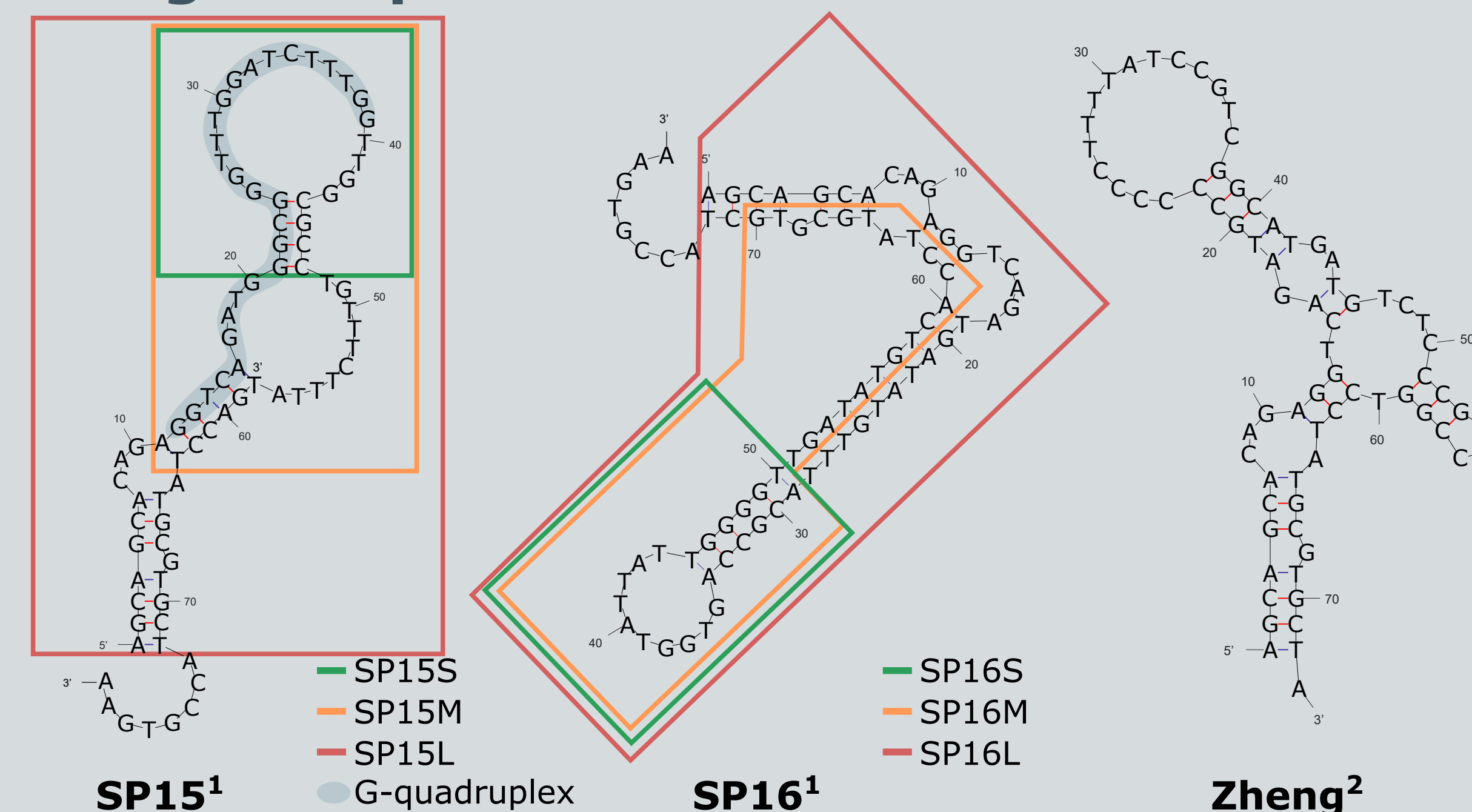
MATERIALS AND METHODS

SPRI principle:

Surface plasmon is an electromagnetic wave at the surface of a thin layer metal - here gold - generated when polarized light hits the surface. At a **specific incidence angle**, the plasmon waves resonate with the incidence light resulting in a **dip of reflectivity**: it is the **resonance peak**.

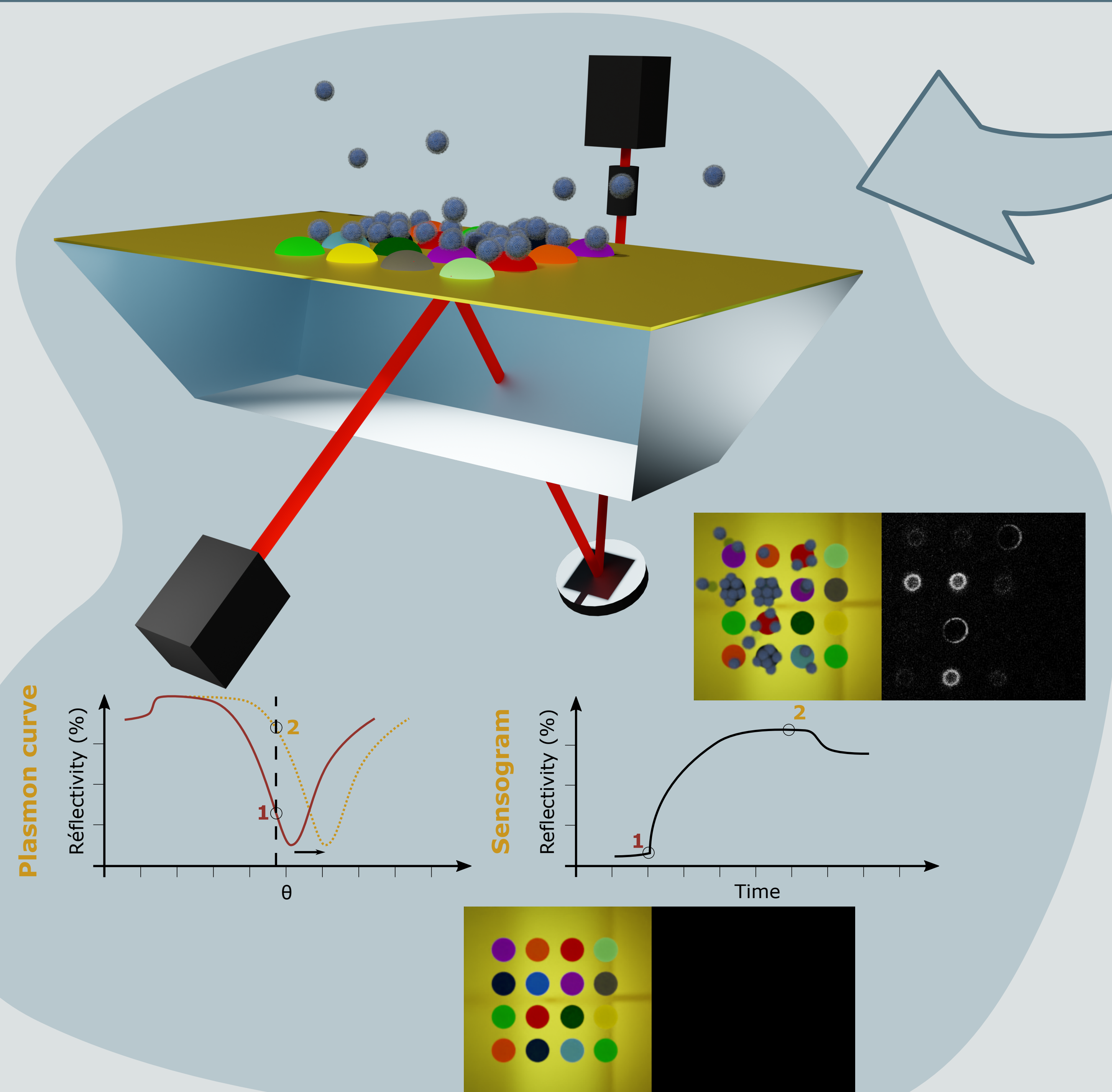
Variations of this thin angle correspond to changes on the biochip surface.

Design of aptamers:



Aptamers from literature^(1,2) were **truncated** according to their **2D structure** and their melting temperature and **stability** using mFold software.

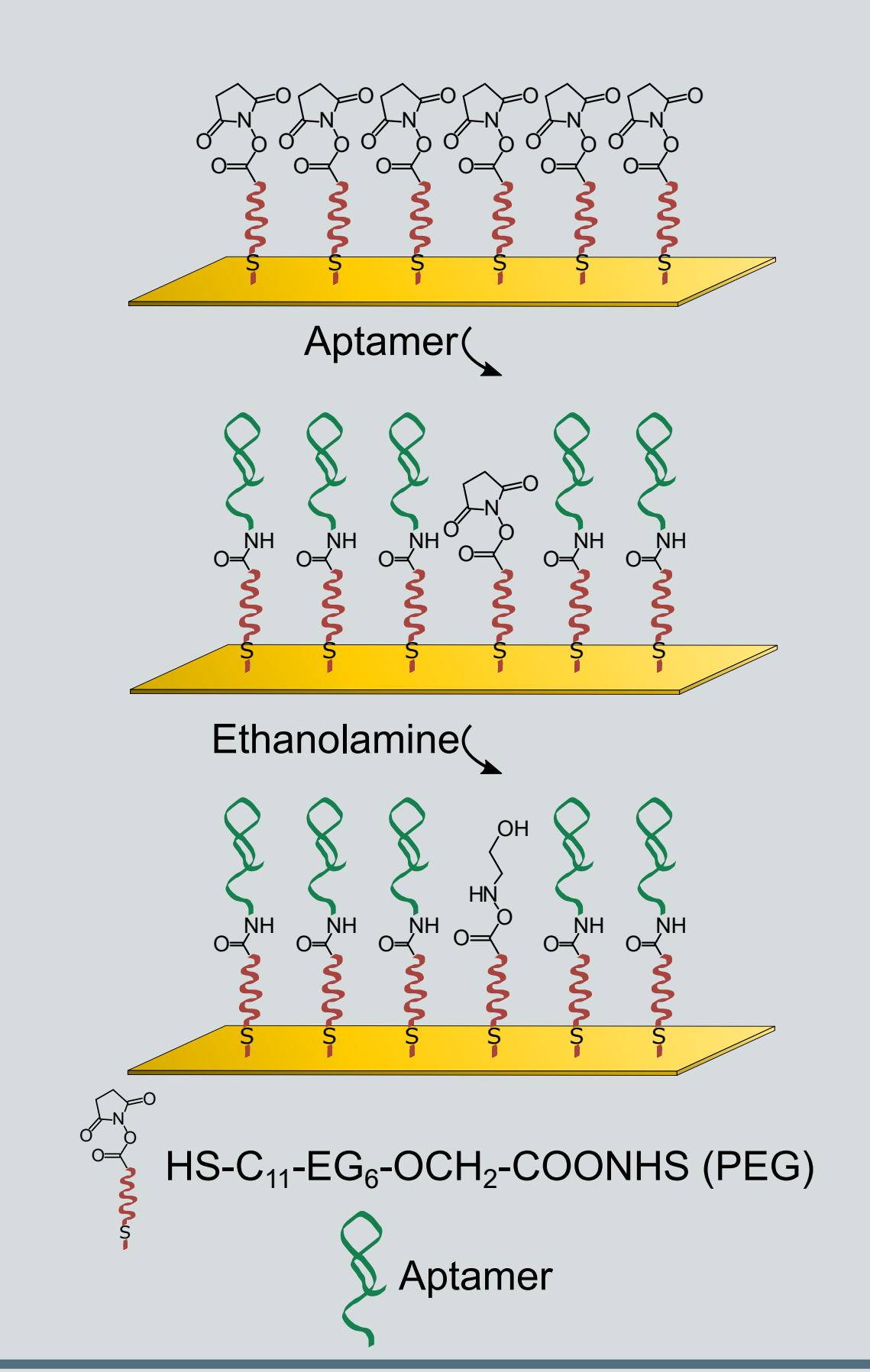
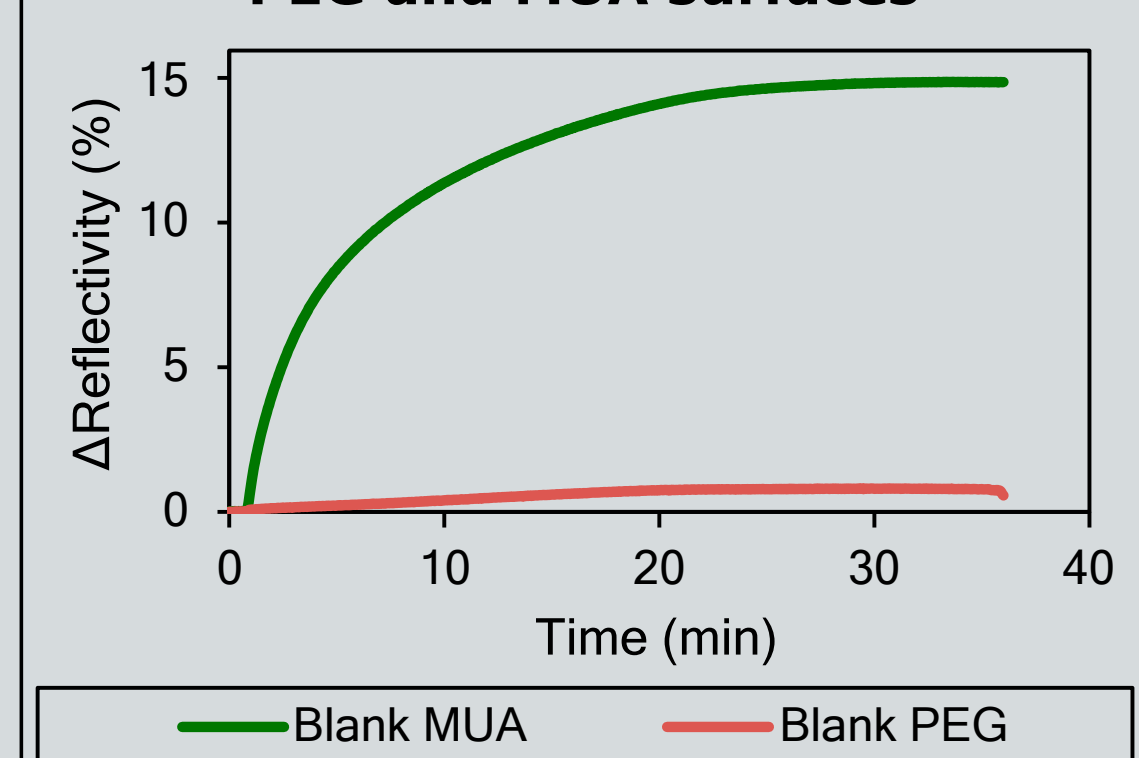
A **5' amine modification** and a **polyThymidine (pT6) spacer** were added to respectively allow aptamer grafting and structuration on gold functionalized surface.



GRAFTING STRATEGY

Two grafting strategies were explored: the EDC/NHS reaction was carried out on either a **11-mercaptoundecanoic acid (MUA)** or **HS-C₁₁-EG₆-OCH₂-COONHS (PEG)** self-assembled monolayer (SAM).

Comparison of the adsorption non specific of *B. cereus* nanosomes on PEG and MUA surfaces

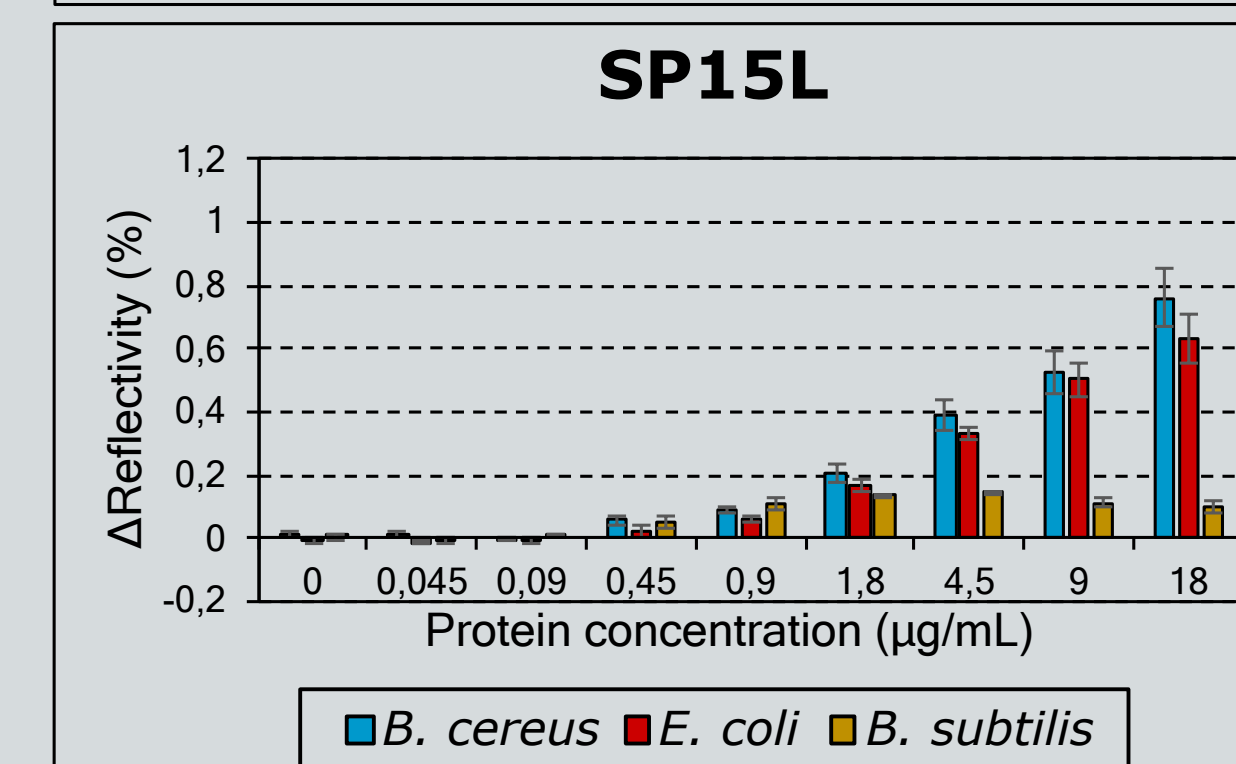
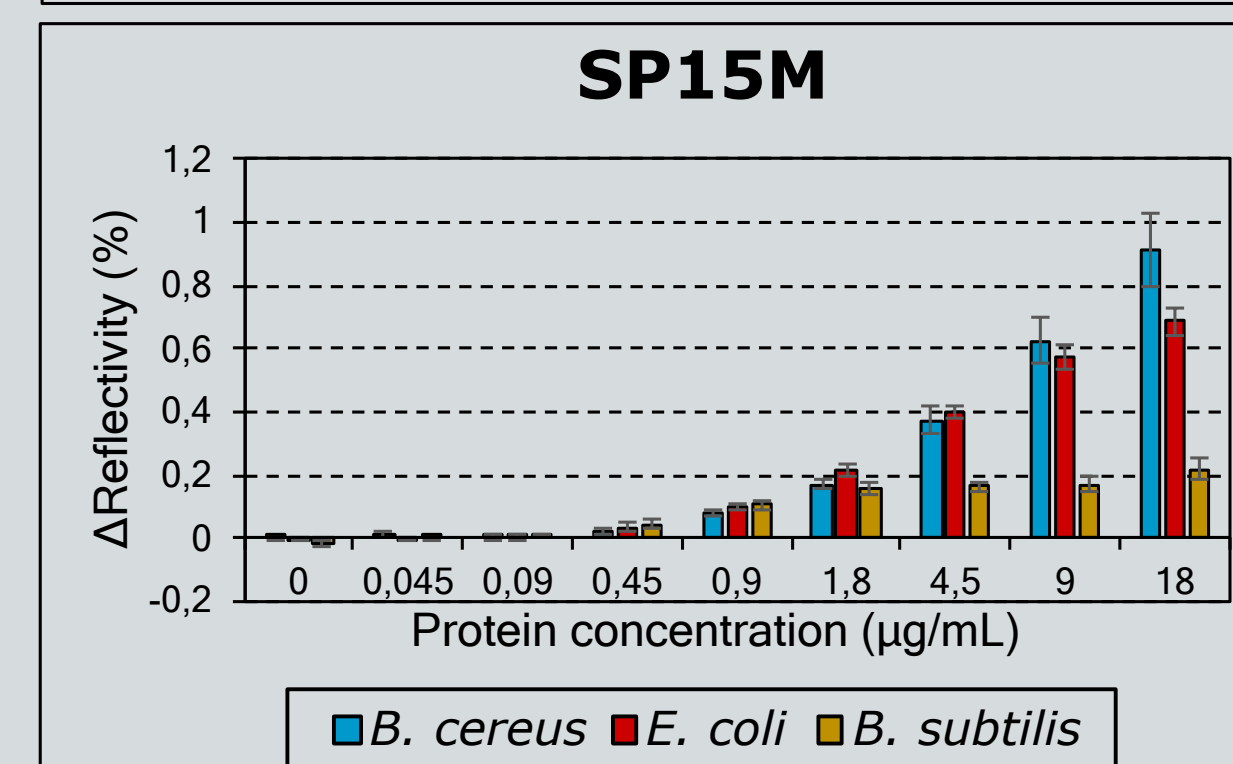
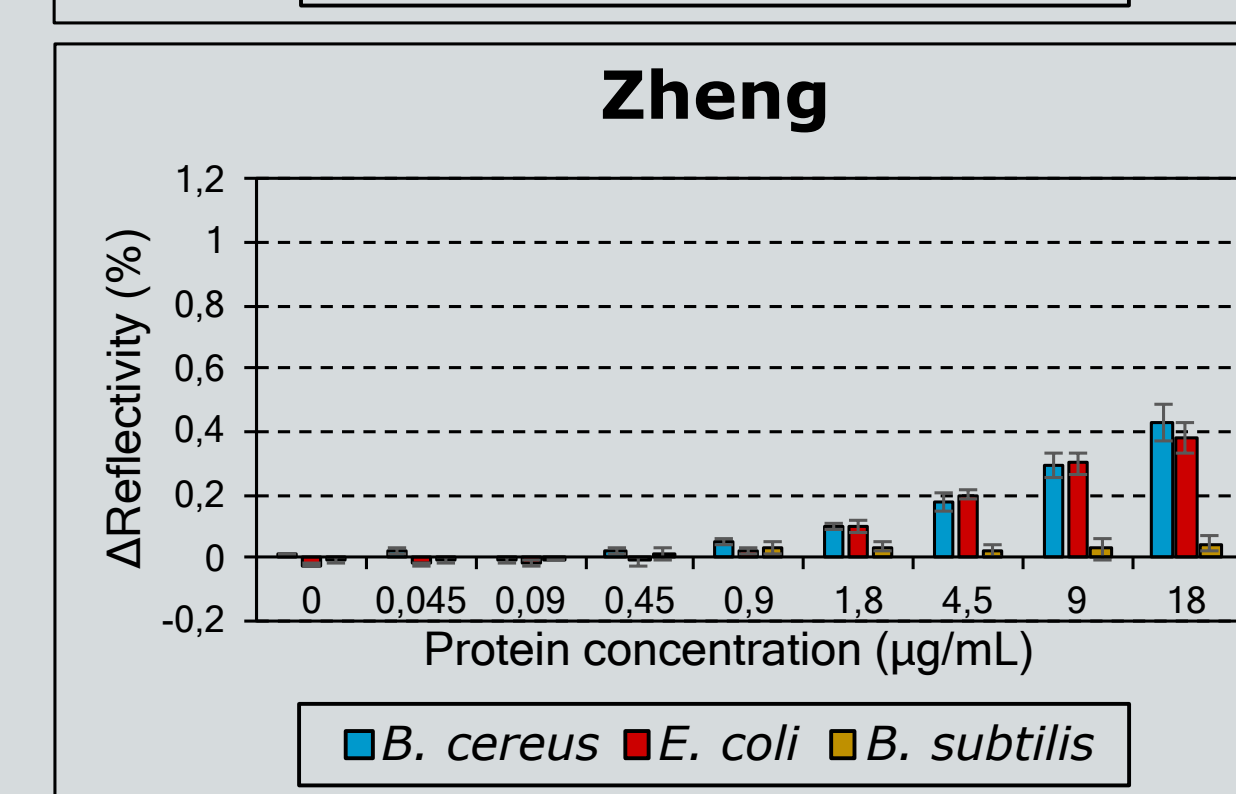
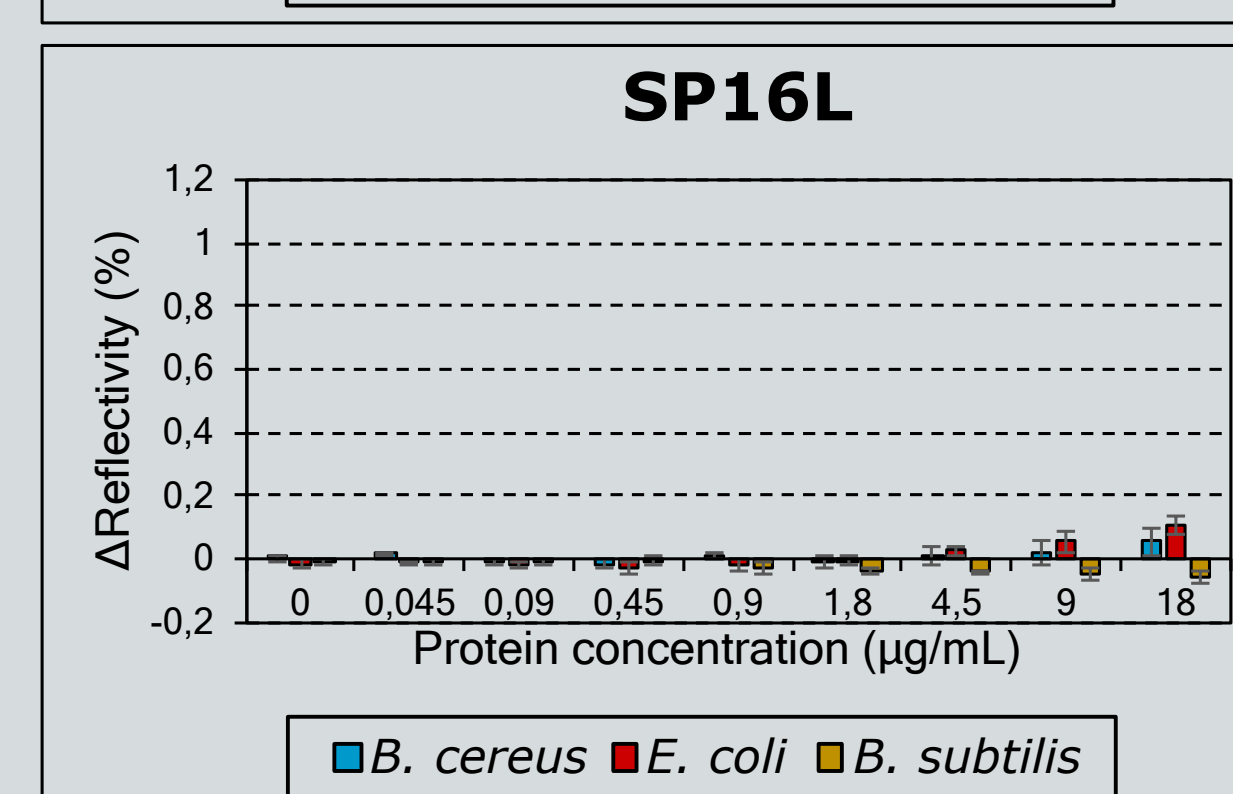
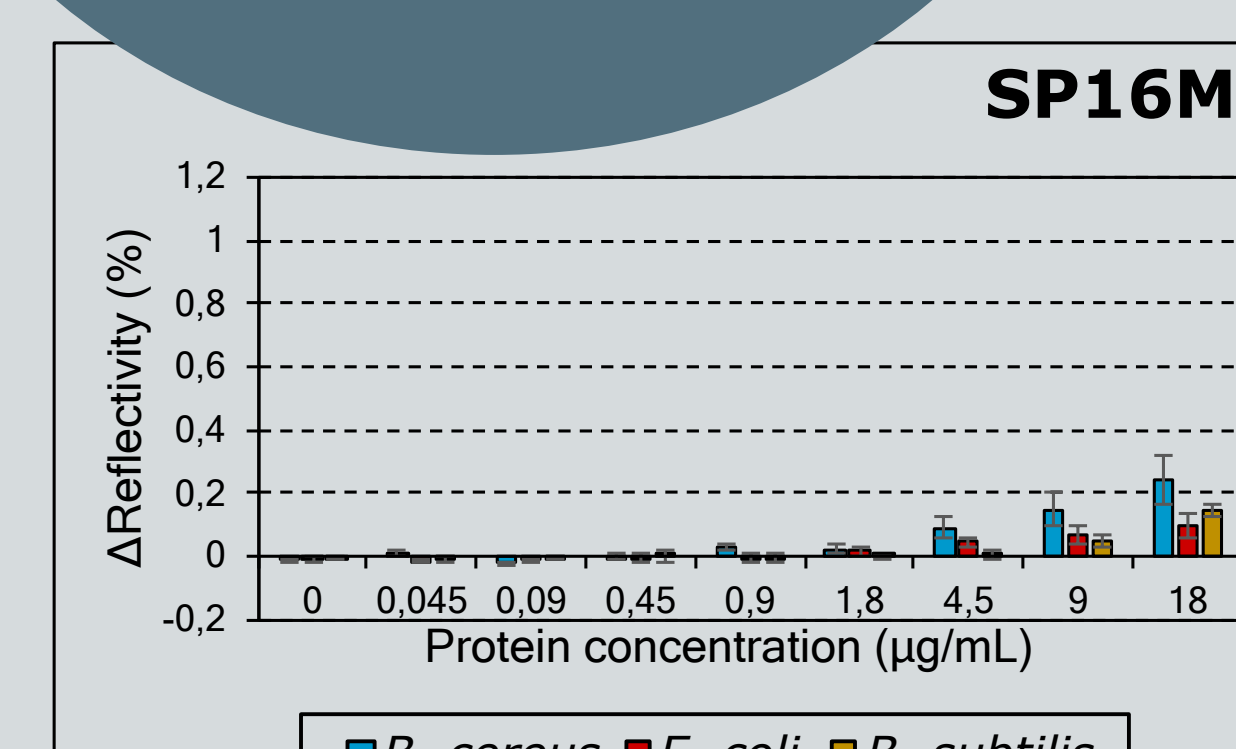
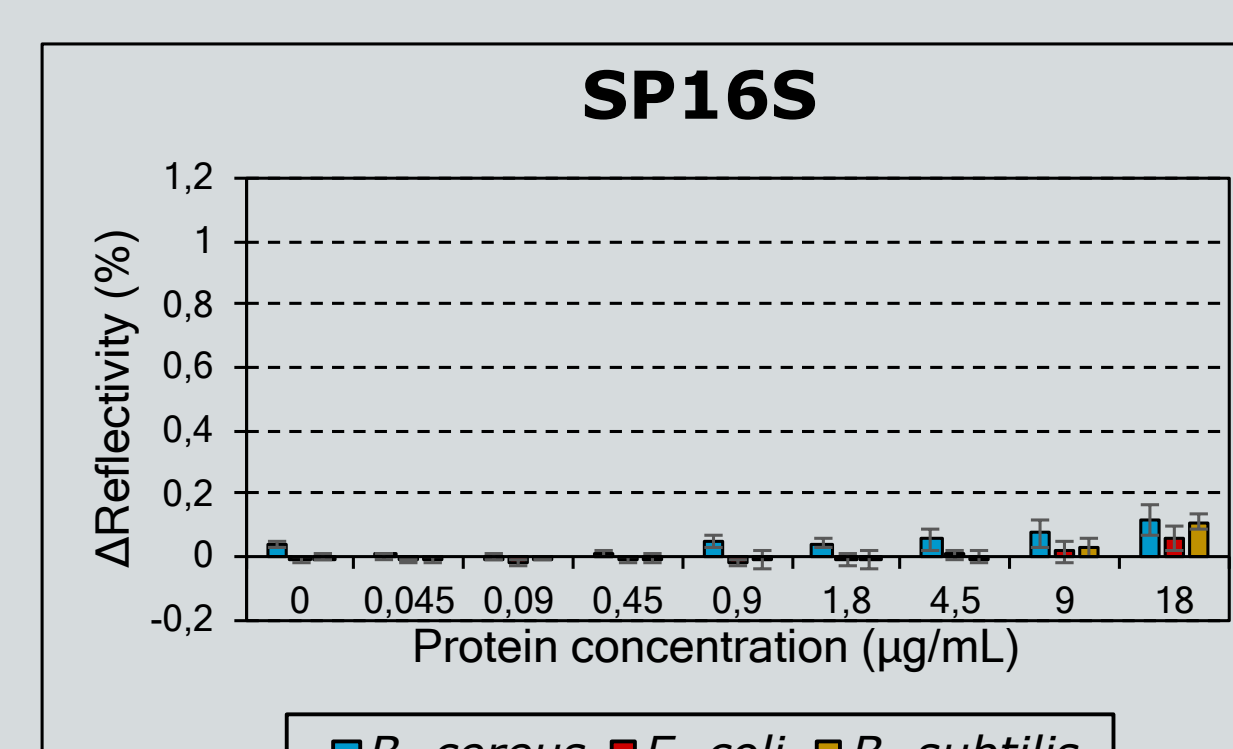
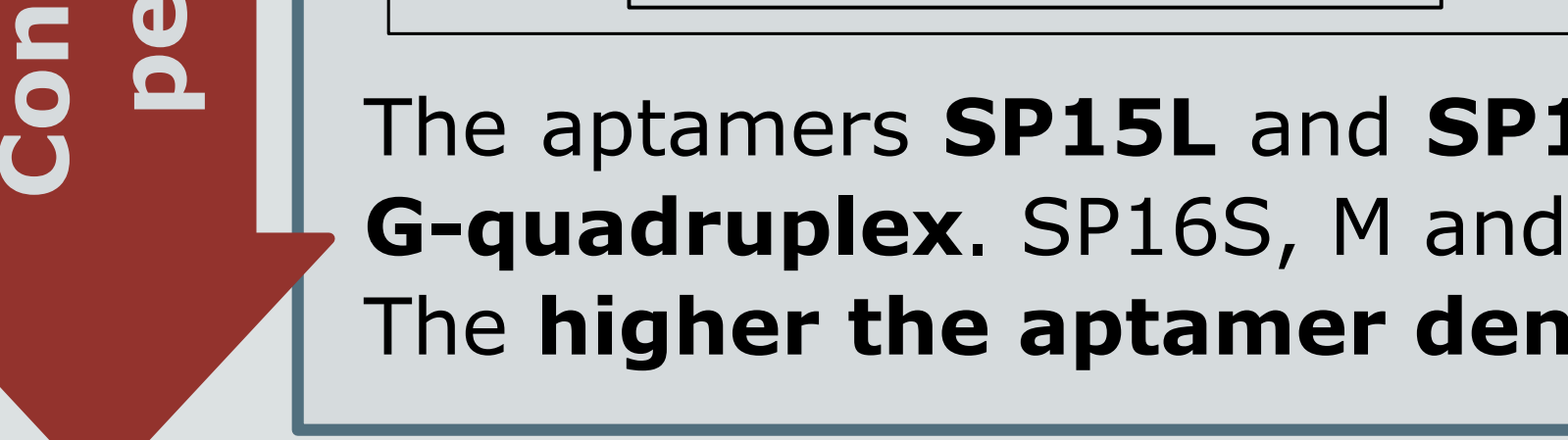
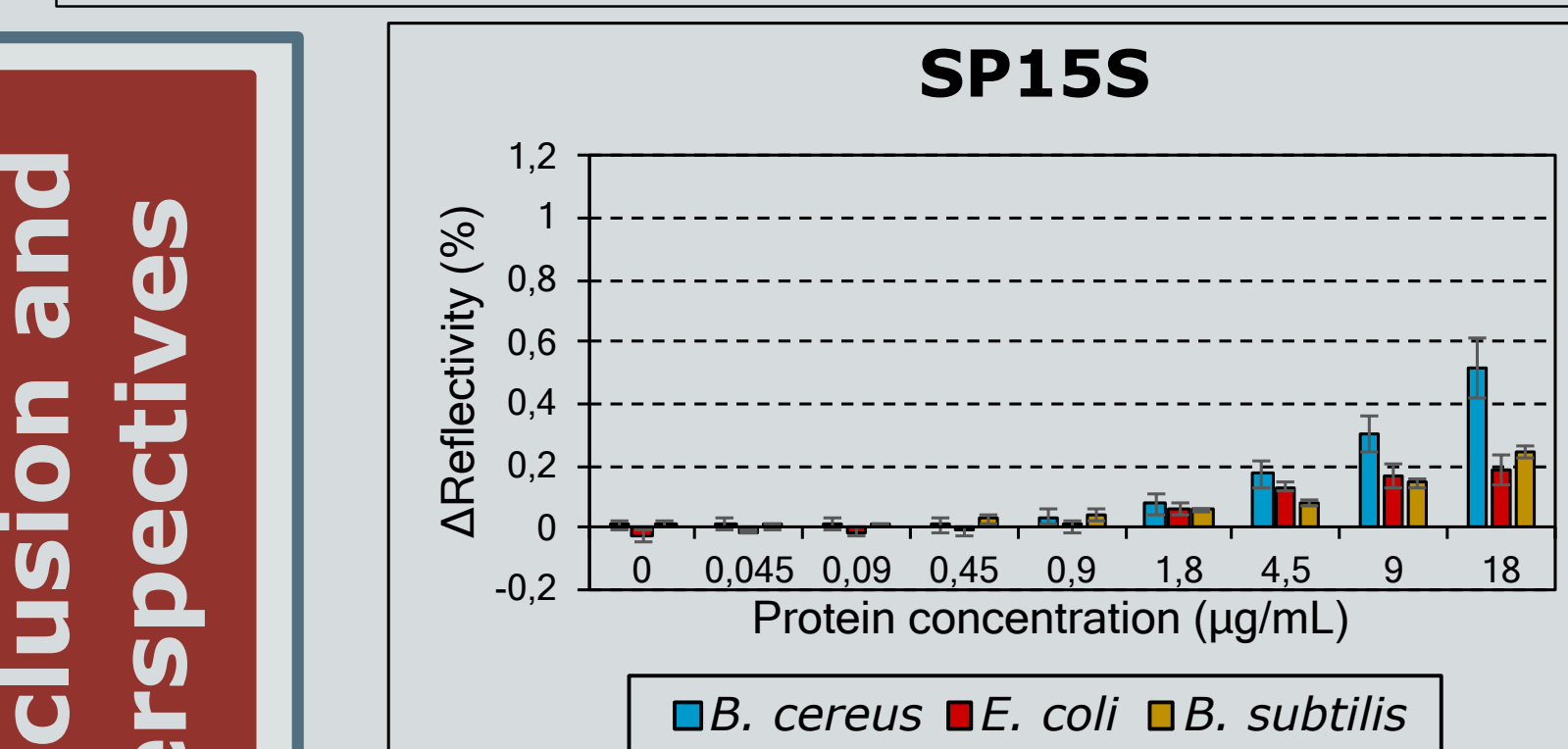
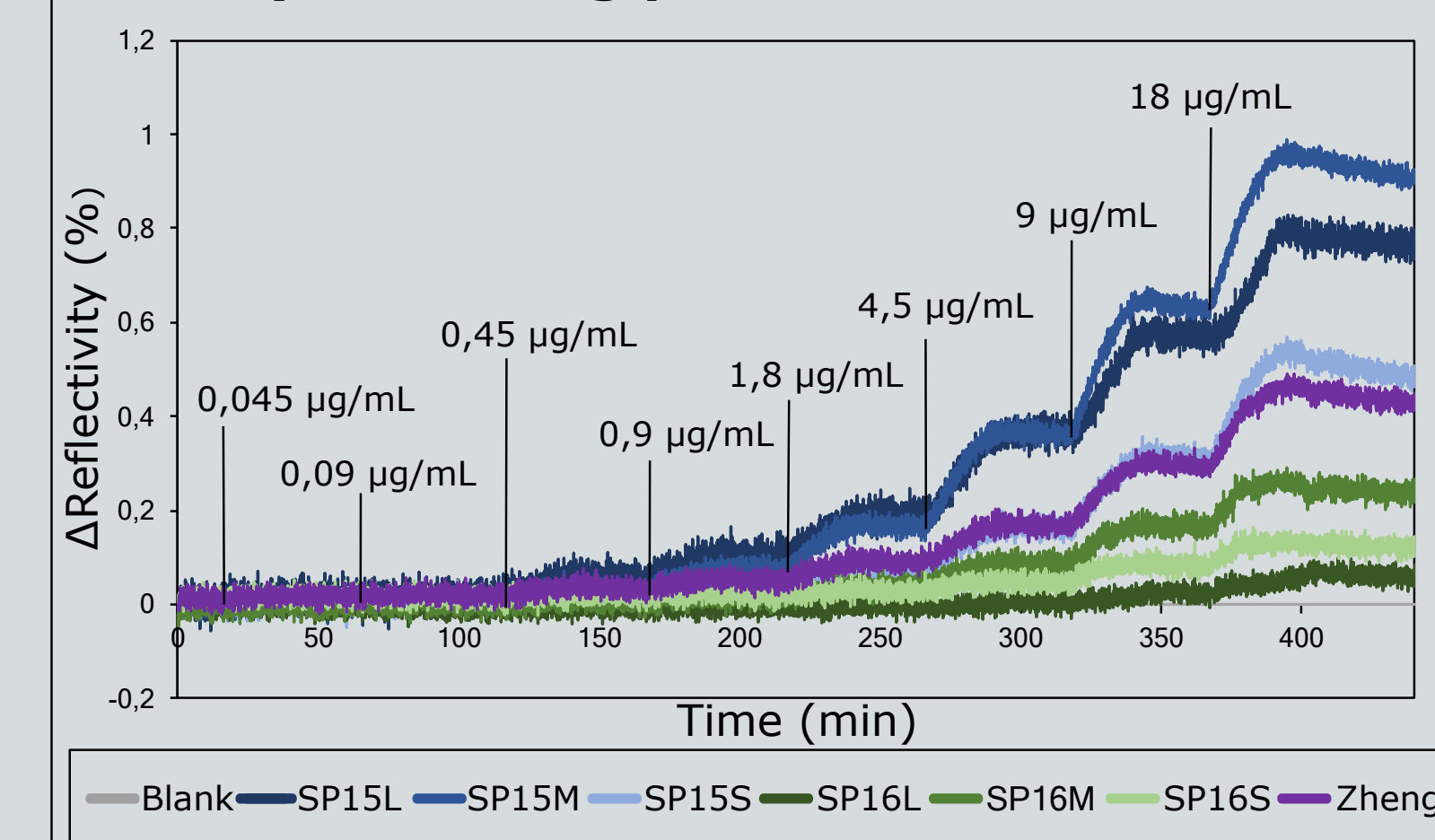


RESULTS

Aptamer **affinity** towards *B. cereus* was explored by **cumulated injections** of its nanosomes.

Specificity is assessed versus *E. coli* and *B. subtilis*.

Cumulated injections of *B. cereus* nanosomes by increasing protein concentration



Conclusion and perspectives

The aptamers **SP15L** and **SP15M** show the **higher affinity** for *B. cereus* and they contain both the **G-quadruplex**. SP16S, M and L have a lower affinity for *B. cereus*. The **higher the aptamer density**, the **better the specificity** seems to be.

The truncated aptamer **SP15M** showed good affinity for *B. cereus* nanosomes. A sequence containing only the **G-quadruplex** will be tested. Nanosomes will be further characterized by **Cryo-TEM** and **NTA**.

