Harnessing the Power of DNA Nanotechnology for Activity-Driven Protein Detection

Alessandro Bertucci
Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Parco Area delle Scienze 17/A, Parma, Italy
alessandro.bertucci@unipr.it

The rapid advancement of DNA nanotechnology has enabled the development of highly controllable, programmable, and versatile nanoscale systems and devices. These systems harness predictable and reversible non-covalent interactions, offering an exceptional platform for molecular information processing. In this talk, we will showcase how DNA nanotechnologies can be engineered to create powerful sensing platforms for specific proteins. Our approach leverages engineered molecular processes that capitalize on the intrinsic biological activity of target proteins, thereby converting their presence into measurable signals.[1] One class of devices we will showcase is dynamic DNA structures that exploit high-affinity protein binding to generate binding-induced measurable signals. For instance, we will discuss the application of structure-switching DNA hairpins capable of detecting and quantifying transcription factors, as well as a structure-switching DNA aptamer targeting the SARS-CoV-2 Spike protein, integrated with carbon nanotube electrodes. [2,3] We will then present a more intricate challenge involving the design of a DNA-based mechanism responsive to the typical proteolytic activity of target proteases. This necessitates the creation of artificial communication between peptide- and DNA-based processes. To address this, we have developed a CRISPR-Cas-powered sensor for an oncogenic matrix metalloproteinase, which utilizes DNA-based amplification of proteolytic cleavage. Our system incorporates a chemical translator, combining a peptide bearing the substrate sequence of the target protease and a peptide nucleic acid (PNA) that converts the protein-based input, i.e., peptide cleavage, into a nucleic acid output. The nucleic acid output can then be processed and amplified. By introducing a rationally designed single-stranded DNA anchored to the PNA sequence of the translator, we can activate the nuclease trans-cleavage activity of a CRISPR-Cas12a system, resulting in the degradation of FRET-labeled DNA reporters. This process generates an amplified fluorescence signal. Remarkably, our strategy enables the detection of the protease MMP2 in the low picomolar range based on its enzymatic activity, surpassing the limits of detection of commercial peptide-based kits by several orders of magnitude. The implications of these studies extend beyond their immediate applications, highlighting the potential of engineered protein-responsive DNA nanotechnologies for transformative advancements in domains such as precision diagnostics and functional synthetic biology.

References

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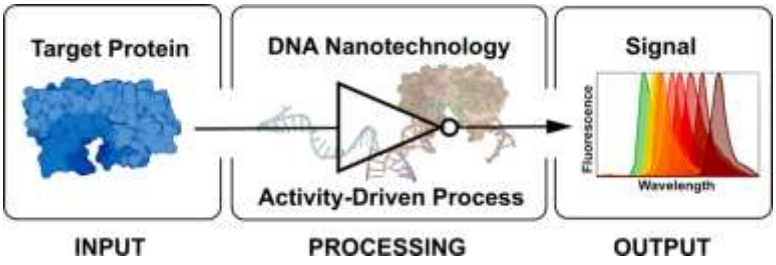

Figure 1: Engineered DNA nanotechnologies can convert protein biological activity into a measurable output.