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CRISPR/Cas-enabled paper-based sensors for rapid monitoring of antimicrobial resistance

CONTEXT and AIM

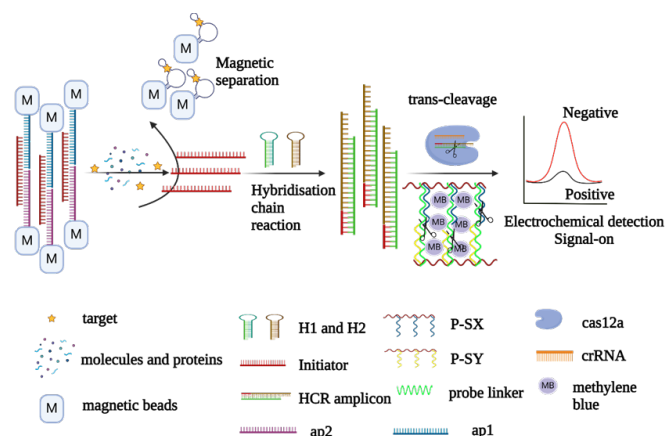
Environmental contamination with pathogenic bacteria and chemical pollutants is a global issue. However, current analytical methods for environmental samples are challenged by the complexity and heterogeneity of the matrices, as well as the ultra-low concentrations of the analytes.

Beyond its extraordinary genome editing ability, clustered regularly interspaced short palindromic repeats (CRISPR) and CRISPR-associated systems (CRISPR/Cas) have recently initiated a new era of biosensing applications. Their high base resolution and isothermal signal amplification provide ultrasensitive, single-molecule level and highly specific sensing.

This capability can significantly improve the detection of low-level contaminants such as antibiotics (including β -lactams etc) in wastewater samples and aid in interpreting the detection with the engineering method onto devices for point-of-need monitoring (e.g. paper-microfluidic analytical devices).

The aim of this work was to develop a novel, ultra-sensitive and low-cost sensing platform to identify chemical pollutants in the environment. This in turn will leverage the CRISPR/Cas platform to offer a range of next-generation environmental sensors for rapid and on-site monitoring of chemical contaminants.

Environmental contamination is a silent crisis threatening ecosystems and human health worldwide. Traditional methods for detecting pollutants are often too slow, costly, or impractical for real-world monitoring". By harnessing the unparalleled precision of CRISPR/Cas systems alongside adaptive bio-recognition elements, we aim to redefine environmental sensing" – Zhugen Yang, Professor of Biosensing and Environmental Health



OUTCOME

A hydrogel-based HCR-CRISPR/Cas bioassay was designed for kanamycin detection. More specifically, we designed a signal amplification strategy to enhance the signal of antibiotics using an aptamer to specifically target kanamycin. The hydrogels wrapping methylene blue particles were synthesised and characterised through square wave voltammetry (SQW), which can be used as redox marker for electrochemical detection of antibiotics. We spent a long time understanding the binding between the selected aptamer and antibiotics using simulation, and we found it is important to identify the high affinity aptamer against the antibiotics.

The next tasks are to optimise the CRISPR/Cas system for signal amplification, and further evaluate the limit of detection, then use the optimised assay to detect antibiotics in real samples.

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