CRISPR/Cas12a based portable and low-cost Inkjet printed platform for one-pot pathogen detection

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Infectious diseases associated to pathogenic bacteria are spreading worldwide, particularly in developing countries. The rapid diffusion of infectious disease urges for rapid and portable devices for the detection of pathogens. Biosensors, because of their low-cost, portability, ease of use and availability for point-of-care/need applications, represent a valid alternative to traditional techniques, that are instead time consuming, expensive and do not allow for a point-of-need analysis [1]. In this work we present an electrochemical portable, sensitive and specific biosensor device for the detection of pathogens. A low-cost platform is fabricated using inkjet printing technology on a plastic substrate using Gold and Silver nanoparticles-based inks and it includes electrode and microfluidic channels. The pathogen DNA target is detected by coupling the recombinase polymerase isothermal amplification (RPA) with the CRISPR/Cas12a system. This last is specifically designed and integrated into the inkjet printed platform. The trans-activity of the CRISPR/Cas12a system triggered after the specific DNA recognition process is used to electrochemically transduce the information by using a smartphone readout. In particular, a rationally designed ssDNA reporter labelled with methylene blue (ssDNA-MB) is functionalized on the working electrode surface and detected by using square wave voltammetry (SWV) electrochemical technique. The trans-activity is triggered when the CRISPR/Cas12a system detects the pathogen target sequence and the reporter ssDNA is cleaved; as a result, the SWV MB associated signal decreases [2]. The performances of the biosensor are assessed and the use of this device for point-of-need applications in real clinical and drinkable water samples are evaluated.

References

[1] Bonini A., Carota A.G., Poma N., Vivaldi F.M., Biagini D., Bottai D., Lenzi A., Tavanti A., Di Francesco F., Lomonaco T., Biosensors, 12 (2022) 894
[2] Bonini, A., Poma N., Vivaldi F.M., Kirchhain A., Salvo P., Bottai D., Tavanti A., Di Francesco F., J Pharm Biomed Anal, 192 (2021) 113645