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AN ENZYME-LINKED OLIGONUCLEOTIDE ARRAY FOR THE ELECTROCHEMICAL DETECTION OF AFLATOXIN B₁

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Mycotoxins are a problematic and toxic group of small organic molecules that are produced as secondary metabolites by several fungal species that colonise crops. With wide ranging structural diversity of mycotoxins, severe toxic effect caused by these molecules and their high chemical stability, the requirement for robust and effective detection methods is clear [1]. The most relevant group of mycotoxins is that of aflatoxins, carcinogenic products belonging to the *Flavus*, *Parasiticus* and *Nomius* species of the genus *Aspergillus* [2]; among these, aflatoxin B₁ (AFB₁) is a potent human carcinogen (first hazard class in accordance with the classification of the International Agency for Research on Cancer) [3].

In this work, an electrochemical enzyme-linked oligonucleotide array to achieve simple and rapid multidetection of aflatoxin B₁ (AFB₁) is presented. The assay is based on a competitive format and disposable screen-printed cells (SPCs). Firstly, the electrodeposition of poly(aniline-anthranilic acid) copolymer (PANI-PAA) on graphite screen-printed working electrodes was performed by means of cyclic voltammetry (CV). Aflatoxin B₁ conjugated with bovine serum albumin (AFB₁-BSA) was then immobilized by covalent binding on PANI-PAA copolymer. After performing the affinity reaction between AFB₁ and the biotinylated DNA-aptamer (apt-BIO), the solution was dropped on the modified SPCs and the competition was carried out. The biotinylated complexes formed onto the sensor surface were coupled with a streptavidin-alkaline phosphatase conjugate. 1-naphthyl-phosphate was used as enzymatic substrate; the electroactive product was detected by differential pulse voltammetry (DPV). The response of the enzyme-linked oligonucleotide assay was signal-off, according to the competitive format. A dose-response curve was obtained between 0.1 ng/mL and 10 ng/mL with a limit of detection of 0.086 ng/mL. Finally, preliminary experiments in maize flour samples spiked with AFB₁ were also performed. From the obtained results, the developed analytical tool has proven itself to be applicable for screening field analysis.

References

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