

Electrochemical Biosensor Arrays

SIMULTANEOUS DNA AND PROTEIN DETECTION

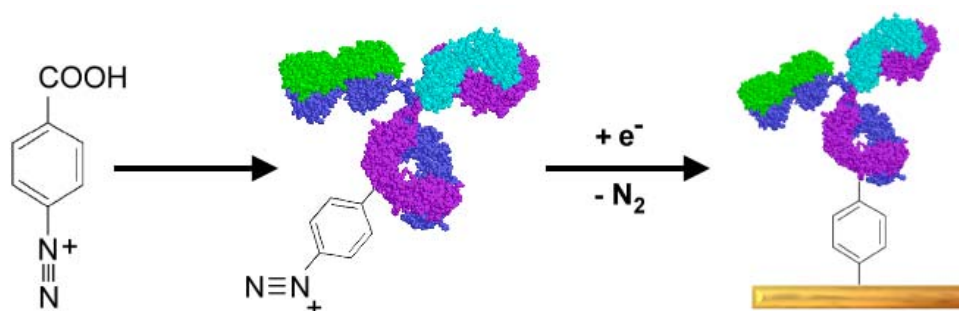
Overview

The reliable and definitive detection of multiple biowarfare agents on a single robust platform would be a significant asset for the defense of our nation and the safeguarding of warfighters. Multiple signature based biosensors can meet this need as they not only allow for multianalyte detection, but also substantially increase confidence in the sensor output as whole cell, genomic, and proteomic data can be interrogated for each target analyte. We have developed a method that allows for controlled and selective immobilization of biorecognition elements onto electrodes allowing simultaneous multianalyte detection of DNA and proteins.

Approach

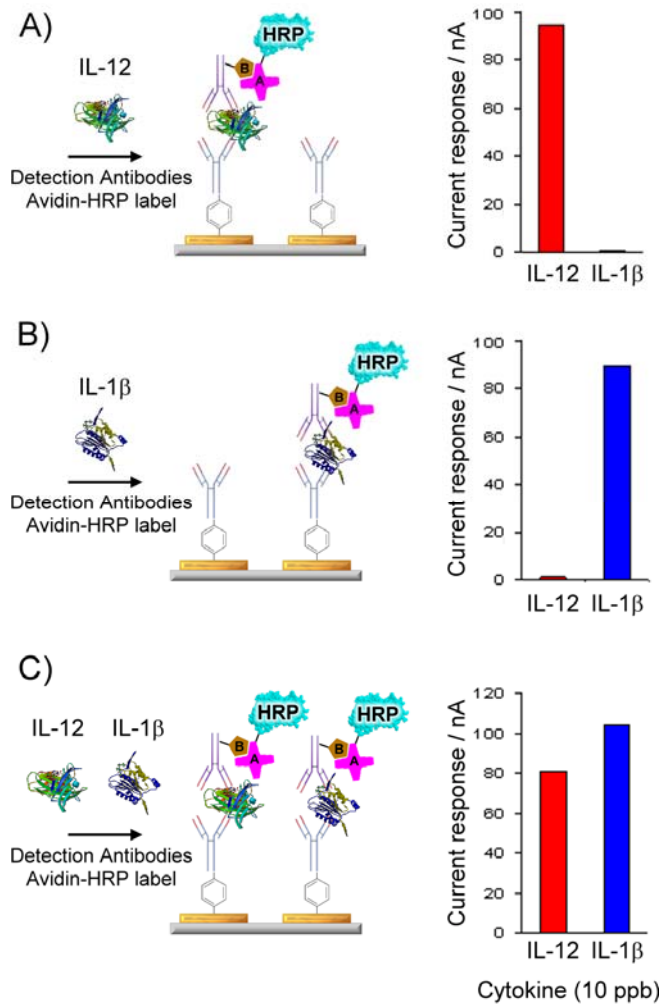
Selective immobilization of biomolecules onto electrodes was obtained using aryl diazonium salts. This patternable surface chemistry forms a highly stable covalent bond with semiconductive or conductive substrates. We have shown the selective functionalization of glassy carbon electrodes with diazonium-horseradish peroxidase conjugates [1] providing facile direct electron transfer and electrochemical detection of peroxide. We have also recently reported the control over surface functionalization density, electron transfer kinetics, and catalytic NADH detection using diazonium modified gold electrodes [2].

Modification of antibodies with diazonium chemistry allows for the selective electrodeposition of antibodies on closely-spaced microelectrodes for multi-target protein detection in an array format, as shown below.



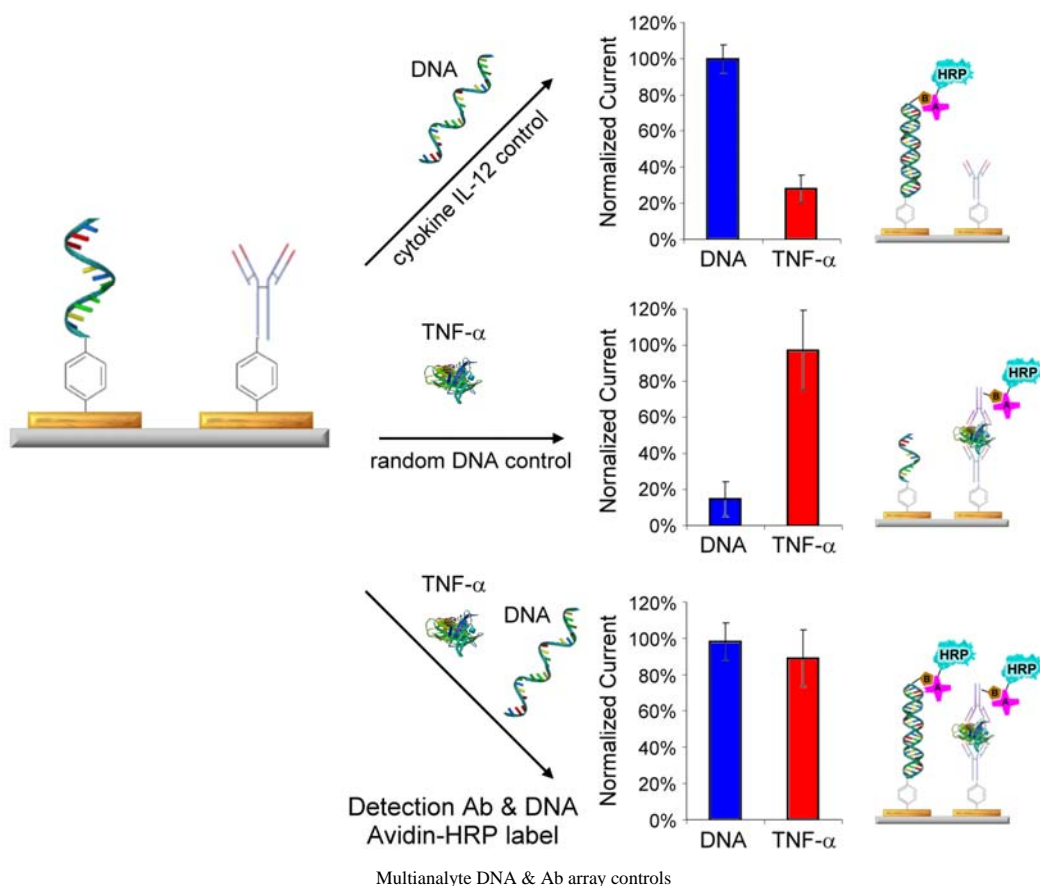
Antibody Mod Scheme

Using this method we have demonstrated simultaneous detection of three different cytokine proteins on a three element array selectively modified with three different diazonium functionalized antibodies [3]. Control experiments, shown below, used a two element array selectively patterned with two different antibodies. These results show the ability to selectively detect either target with minimum cross-talk between electrodes.



Multianalyte Immunoassay Controls

We have also demonstrated the selective immobilization of DNA probes and antibodies on a 9 element electrode array using diazonium chemistry. The figure below shows the simultaneous electrochemical detection of DNA and protein on a single platform. (Gold array with 5 electrodes patterned with DNA and 4 with anti-TNF-a antibodies)



References:

1. **“Diazonium-Functionalized Horseradish Peroxidase Immobilized via Addressable Electrodeposition: Direct Electron Transfer and Electrochemical Detection”** R. Polsky, J. C. Harper, S. M. Dirk, D. C. Arango, D. R. Wheeler, S. M. Brozik, *Langmuir*, 2007, 23, 364-366.
2. **“Electro-Addressable Selective Functionalization of Electrode Arrays: Catalytic NADH Detection Using Aryl Diazonium Modified Gold Electrodes”** J. C. Harper, R. Polsky, S. M. Dirk, D. R. Wheeler, S. M. Brozik, *Electroanalysis*, accepted.

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