Ph.D. Thesis

Advanced Electrochemical Biosensors for Low-Molecular Weight Analytes: Integrated Tailorable Strategies for Sensitive and Selective Detection

Abstract

The current environmental and health concerns associated with chemical, or pathogen exposure have led to an increasing demand for novel, sensitive detection technologies which can be readily adapted to point-of-care devices. Biosensing, in particular, has gained attention from the scientific community for its remarkable adaptability and increased analytical performance. Within this category, electrochemical-based biosensors have shown great potential for field applications where low analyte limits of detection are required. Moreover, the combination between this transduction method and short biological or synthetic receptors (like aptamers or biomimetic molecules) has delivered great analytical results with high sensitivity and specificity towards low molecular-weight analytes.

In this thesis we explore the development of highly tailorable electrochemical biosensors, utilizing oligonucleotide aptamers as capture probes for small molecules and we propose the rational development of novel biomimetic molecules with great potential for sensing applications. The main goal of the work (i.e. the design, preparation and evaluation of the analytical response of biosensing platforms for low-molecular-weight analytes) has been achieved though sequential analysis of favorable development strategies that would tackle the main challenges of sensor fabrication.

Firstly, we have investigated the most suitable carbon substrate for biosensor preparation. We have performed morphological, chemical and electrochemical analyses on two types of glassy carbon electrodes modified with electrochemically reduced graphene oxide and have investigated the effects of the deposition methodology on the surface coverage and architecture. This study provided the first comparative assessment of such type, and it provides crucial information when selecting the most appropriate modification method.

Secondly, we have proposed two original methods for the development of controlled surface-functionalized carbon-based platforms with tailorable sensitivity and antifouling properties, starting from well-established strategies, and extending the current knowledge to provide facile methodologies for advanced sensor fabrication. We have looked at the effect of biomolecular receptor surface density on sensor performance, and we have expanded the current practice of electrografting of silyl-protected aryldiazonium salts to include bulkier protective groups, as a strategy to control the biomolecule surface distribution. Taking good note of one of the main challenges of real sample analysis, namely matrix fouling effects, we have proposed a facile way to co-immobilize antifouling molecules and bioreceptors with precise ratios between the two

elements. These studies have proven that the proposed strategies for controllable sensor fabrication deliver excellent analytical results, while tackling real sample challenges.

Lastly, we have rationally designed N-substituted polyglycine oligomers (peptoids) with complexation capacities for a model antibiotic molecule, achieving or third objective of the current work, namely the development of a novel type of biomolecular recognition element for low-molecular weight analytes. We have investigated helical peptoid molecules with selected side-chain chemistries that would promote conjugation with our target analyte. Sequence investigations have highlighted the importance of side-chain position and molecule folding for successful complexation.

The current thesis has provided great contributions for biosensing technologies, highlighting an integrated approach for device fabrication. The results obtained not only advance the current scientific knowledge of this field but also provide concrete strategies to tackle controllable sensor design and real sample analysis that can be further expanded to applications in point-of-care detection devices for small molecules.