ABSTRACT

This work focused on developing metal nanomaterials with hydrophobic characteristics to deposite through the Langmuir-Blodgett (LB) film process. It is expected that a monolayer of metallic nanomaterials with enhanced conductivity could provide a better choice for effective transportation of charge and improved biomolecule adsorption capabilities compared to other methods and materials used for developing immunosensor platforms. An immunosensor based on the monoclonal anti-glutamate antibodies immobilization onto a thin film of new nanocomposites of polymer-metallic nanoparticles, conductive polymer-metal oxide nanoparticles, and conductive polymer-metallic nanoparticles through a self-assembly process developed. New platforms developed in this thesis concurrently manage functional groups' availability for antibody immobilization with enhanced electrical conductivity. An antibodies-based immunosensor is ideal for industrial applications since it allows for detecting MSG at concentrations ranging from nano-molar to micromolar on a single platform. A hypothesis for low concentration detection is proposed by considering the following facts;

In-situ reduced chitosan gold nanoparticles matrix provides both higher sensitivity and extended limit of the detection range of MSG. It is expected that impregnation of the in-situ reduced gold nanoparticles in the chitosan network provides better charge transportation and enhanced selective functionalization capability and biocompatibility. The proposed in-situ reduced gold nanoparticles may have better chemical coordination and homogeneous distribution in the chitosan network. Thus, this reduction process has an advantage over the methods that adopt the physical mixing of nanoparticles with polymeric materials. An enhancement of four-fold in current has been achieved by incorporating GNP in chitosan. The formulation also has the advantage of stable and adhesive coatings on working electrodes to develop a platform based on monoclonal anti-glutamate antibodies immobilization, Coating of these nanomaterials allowed to achieve the highest detection

sensitivity with the lowest detection limit of 0.1 nM. Electrochemical characteristics of chitosan-gold nanoparticles were studied by measuring relative change in redox current compared to standard working electrodes. Antibodies' interaction with the CS-GNP platform was confirmed through a reduction in electrochemical current.

The second method uses the titanium dioxide (TiO_2) nanoparticles in the PANI matrix and optimizes conditions for preparing LB films of the polyaniline-TiO_2 nanocomposite. A unique strategy was adopted in which electrochemical polymerization of aniline in the presence of TiO_2 nanoparticles was performed on ITO substrate, dissolving it in a solution of NMP and isopropanol, and LB film deposition of contamination-free subphase of PANI-TiO_2. Thus, the work achieved a higher degree of polymerization through the electrochemical process and subsequently used this polymerized material to make LB film. The antibodies immobilized electrodes were successively used to quantify MSG ranging from 1 nM to 500 μM in the standard electrolyte. A linear relationship was obtained between the current change and the MSG concentration.

In the third method, the chemical synthesized of surfactant-free stable ultra-small gold nanoparticles (< 10 nm diameter) in the PANI matrix were used. Electrochemical polymerization of aniline monomers with these gold nanoparticles was performed on ITO substrate. This unique method enabled the synthesis of polyaniline-GNP nanocomposite with hydrophobic nature. Further, this PANI-GNP nanocomposite was dissolved in a solution of NMP and isopropanol, and the conditions were optimized for preparing LB films of the polyaniline-GNP nanocomposite. The developed thin film shows a very high conductivity with improved capability for the adsorption of biomoles. The developed electrochemical biosensor shows stability over a wide range of pH values. Antibodies immobilized immunosensor were sequentially used to quantify MSG ranging from 1 nM to 10 mM in the standard electrolyte and 1 μM to 1 mM in tomato sauce. A linear relationship was obtained between the current change and the MSG concentration.

The electrochemical properties of all the immunosensors mentioned above were investigated by measuring the relative change in redox current compared to standard working electrodes. To improve measurement sensitivity, monoclonal anti-glutamate antibodies were bound to the amine-functionalized spots on the working electrode using the carbodiimide coupling technique. Since monoclonal anti-glutamate antibodies were used due to their ability to bind the uniquely designed substrate and arrange reactive groups in a manner that promotes a specific reaction transition state, they give these antibodies their selectivity. Monoclonal anti-glutamate antibodies show stereoselective properties (e.g., the monoclonal anti-glutamate antibodies used in work reported in this thesis are specific for the L-isomer of glutamate). The developed immunosensors can quantify the concentration of MSG in the nano-molar range with higher stability and reproducibility.

This thesis work will be beneficial for the researcher/ academics / industrial working in the area of electrochemical quantification of monosodium glutamate biosensor