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ELECTROCHEMICAL SENSOR FOR THE DIAGNOSIS OF TRAUMATIC INJURIES OF THE CENTRAL NERVOUS SYSTEM

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INTRODUCTION

Injuries of the central nervous system (CNS) such as traumatic brain injuries (TBI) and traumatic spinal cord injury (tSCI) are widespread. Approximately, 170,000 individuals in Canada, suffers these injuries every year[1]. Majority of the patients are left permanently disabled with limited restorative treatment and the cost of these CNS injuries is approximately $15billion a year to the Canadian economy[2]. One of the ways to accurately diagnose and manage these injuries for the effective outcome is by the detection of Biomarkers. Concentrations of biomarkers are found to be critical and provide vital information of health and healing of tissues. Accurately measuring these can help in assessing the heath and healing of the CNS during injury. We report detecting of S100β, glial specific protein primarily expressed by astrocytes. Median concentration of S100β before injury is approximately 45pg/ml while post injury it increases five folds to 240pg/ml[3]. We report a novel electrochemical biosensor for sensing S100β in the dynamic range of 1pg/ml to 1ng/ml using this biosensor.

METHOD

Electrode was connected to Biologic SP150 potentiostat for Electrografting (Fig.1). Electrografting of the sensor's surface was performed by adding 50µl of 10mM 4-nitrophenyl Diazonium (4-NPDS) solution in PBS and performing Chronoamperometry (CA) followed by washing the electrode with DI water. 0.1M KCl was pipetted on the sensor's surface and Cyclic Voltammetry (CV) was performed to generate nitrophenyl film on it. Electrode surface was washed using PBS and incubated for 60mins in 2.5% Glutaraldehyde (GA) in PBS (200mM pH7.0). After 60mins, 50µl S100β (1% v/v) in PBS was pipetted on it and incubated for 30mins. Electrode was washed with PBS and 100µl of blocking agent (0.25% of BSA in PBS buffer pH7.0) was pipetted and electrode was incubated for another 30mins. This completes the surface modification ready to detect S100β protein (Fig 2). Electrochemical analysis was performed using 100µl of 5mM Potassium Ferricynide $K_3Fe[Cn]_6$ redox pair under Differential Pulse Voltammetry (DPV)[4].

RESULT

Characterization of the electrode was done to ensure that we receive unique current peak for each concentration of the S100 β. Redox current observed for each stage of electrode modification decreased compared to bare electrode (Fig. 2). We were able to detect S100β spiked in PBS in the dynamic range of 1pg/ml to 1ng/ml using electrochemical biosensor (Fig.3). Peak current increased linearly with the increase in the concentration

of S100 β. However, after 1ng/ml detection current remained stable.

CONCLUSION

We have developed a sensor for the electrochemical sensing of CNS injury marker s100 β. We have shown the change in concentrations of the biomarker results in the change in peak current which can serve as method of detection CNS injury.

FIGURES

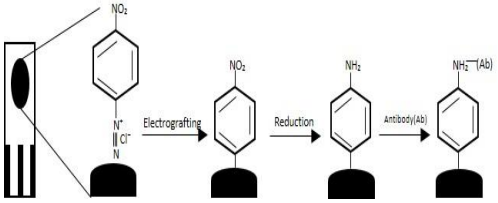

Fig1: Fucntionalization of the electrode with immobilized antibody

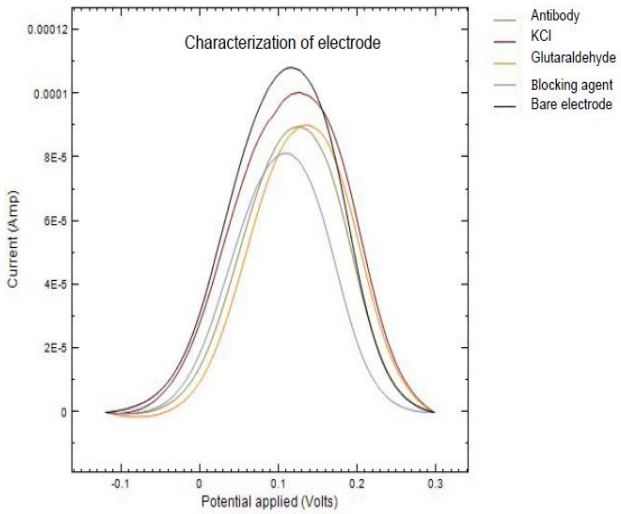

Fig 2. Characterization of the electrode after each stage of surface modification.

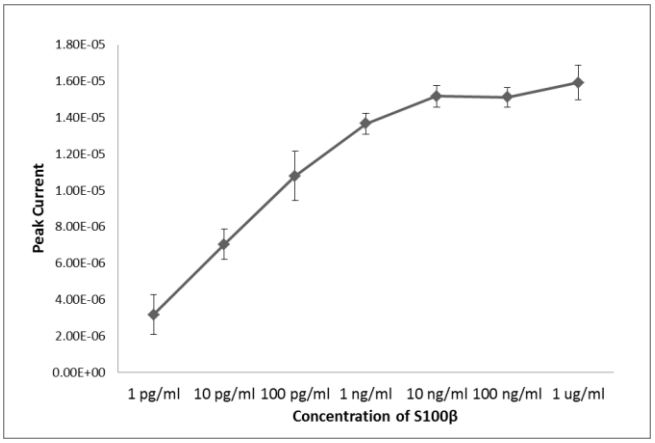

Fig 3: Detection peak current associated with the concentrations of S100β concentration and error bars are standard error (n=3).

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