Graphene Patterned Microchip For Colorectal Cancer Detection

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ABSTRACT

Cancer currently stands as the second-leading cause of death worldwide. Studies reveal colorectal cancer (CRC) to be the 4th leading cause of mortality due to cancer. It is estimated that about 30% of CRC cases are hereditary, of which 5% are attributed by known syndromes, particularly Lynch Syndrome. This pilot study aims to fabricate a DNA-graphene-polypyrrole (DGP) based biosensor to diagnose deficiency of functional MMR proteins present in patients at a scale of less than ng/ml. We have followed LAB-on-CHIP method. We find that the interactive forces between avidin and graphene are mainly hydrophobic, along with some van der Waals, electrostatic and hydrogen bonding interactions. Different mismatch combinations were performed, to prove the activity of each component on the chip. 30 such combinations were done. Electrochemical impedance spectroscopy was done to confirm the working of the bio sensor by corresponding change in electrical impedance. To assist this real-world system, we have carried out simulation studies as well. In the simulation studies from 0-200ns, we present the progression structures of human MutS protein to biotinylated DNA that has been fixed to simulate the manner of a biosensor, furthermore the mismatch within the DNA has been manually introduced with the aid of computational tools to reveal the interactions of the DNA and the protein. This research additionally permits us in early detection of colorectal cancer and the mapping and expertise of the method related to the area of the mismatch repair.

EXPERIMENTAL METHODS AND RESULTS

- Chromium based nickel plated chips were used to fabricate DNA-Graphene-Polypyrrole (DGP) based biosensor.
- Gppb substrate was formed using cyclic voltammetry set up with initial conditions 800mV final 900mV at a scan rate of 20mV/s for 100 cycles.
- Avidin was deposited and impedance was measured using Electrochemical spectroscopy.
- MMA dilutions were performed, around 27-28nanomoles of DNA was poured on the dry chip and EIS was measured.
- Similar step was repeated with MutS protein.
- Concentration of DNA and MutS was decreased with each trail and the system was measured was measured for least possible concentration.
- Impedance of different combinations of the five components involved in fabrication of biosensor was measured. Around 40 combinations were performed.

SIMULATION STUDIES

- The system was modeled using VMD⁴ and the simulations were carried out foe 200ns using NAMD⁵.
- CHARMM force field was used for NAMD simulations.
- TIP3 water model was used for solvation and further ionization of the system.
- The mismatched DNA segment (50/51 bp) was generated using “Random DNA sequence generator” tool.
- The DNA was fixed at the CYT at the 5’ end specifically at the atoms 1641 to 1668.
- VMD plugins for RMSD, H2 bonds and Interaction energy are used along with the TCL scripts for Center of Mass variation and Number of atoms.

REFERENCES

2. A. Amruini, D. Thierry * Swedish Corrosion Institute, Roslagsvigen 101 Hus 25, S-104 05 Stockholm, Sweden, Application of electrochemical impedance spectroscopy to study the degradation of polymer-coated metals.