Bio-molecular impedance biosensing in Nanofluidic devices

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Abstract

A small chip for biomedical analysis connected to our home computer or smart mobile, which would be capable of diagnosing illnesses, a lack of vitamins, or the over-presence of substances from samples of blood, urine or saliva would be a great advance. Such a system could give advice to the user about the optimal medicines to take or provide information to a specialist for effective treatment. Of course this system will take some time to develop but this thesis aims to provide some key understandings to help make such systems a reality and bring some new biosensing elements to this exciting project by investigating the molecular sensing of proteins in well-defined nanometer-sized confined areas. The understanding of molecular dynamics in nano-confined volumes is fundamental for designing the appropriate lab-on-a-chip devices able to transport and sense biomolecules. However, the advantages and problems occurring at the nanoscale are still to be discovered and currently, there is a lack of accurate sensing devices for proteins in nanofluidics. One limitation for performing these studies and biosensing device developments is to have a low-cost and simple nanopore biosensing platform. To address these limitations this thesis focuses on exploration on nanoporous alumina (NPA) with perfectly ordered nanoporous or nanochannels prepared by unique self-ordering electrochemical process with the aim of developing new nanofluidic biosensing platform with new functionalities that are not accessible to microfluidics. Based on measurements performed in 20-70 nm nanochannels, where proteins were binding on the internal surface of the nanochannel and its interactions with antigens were investigated using electrochemical impedance spectroscopy measurements. The size of prepared nanofluidic channels is comparable to the length scale for electrostatic interactions in aqueous solutions

and the binding of proteins in the various dimensions and shape of nanochannels were modeled theoretically and verified experimentally with impedance spectroscopy. As a result of electrostatic interactions, surface charge can govern ionic concentrations in nanofluidic channels. On the other hand, it has been shown that protein charges directly influence the nanochannel conductance giving a better understanding of how the protein's counter-ions modify the surface charges inside the nanochannels. A direct measurement inside the nanochannels has allowed the identification of different systems of interacting proteins, depending on the thickness of the electrical double layer. Due to the small channel size, surface binding of protein and a generated electrostatic conduction effect inside the nanochannel due to the charge of the proteins and ionic strength of the solution have important role in the impedance biosensing. An understanding of the properties and advantages of the nanoporous alumina nanochannels lead to the various other applications including the extraction of DNA and proteins, and measurement of the activities of bacterial nanowires. Finally, a novel microchip biosensing device with an NPA platform is designed and demonstrated for impedance biosensing to measure the changes inside the nanochannel due to the binding of proteins. The results showed that changes in the impedance can indicate target binding and sample surface morphology is responsible for changes in the sensing ability of the developed device.

The work described in this thesis details significant research in the nascent field of nanofluidic biosensing. The work points out novel, important, experimentally-verified complements to define theoretical models as well as practical approach to go forward with the design of complex nanofluidic systems applied to biomedical and biological applications.

Declaration

'I certify that this thesis does not incorporate without acknowledgment any material previously submitted for a degree or diploma in any university; and that to the best of my knowledge and belief it does not contain any material previously published or written by another person except where due reference is made in the text'.

Krishna Kant

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The source of this life is the Mystery that I do not understand. To that One I owe everything.

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List of Publications

Invited Book Chapter

1. **K. Kant**, D. Losic, Focused Ion Beam (FIB) technology for micro and nanoscale fabrications Book: "*Lecture notes in Nanoscale Science and Nanotechnology*", Eds. Z.M. Wang, A. Wang, G. Salamo, K. Kishimito, S. Belluci, Y.I. Park, Springer, 2013. (Invited)

Refereed journal publications

1. **K. Kant**, C. Priest, J. G. Shapter, D. Losic, "*Microbial cell lysis and nucleic acid extraction through focused ion beam milled nanofluidic channel*" Lab on Chip, 2014. (Under review)
2. **K. Kant**, C. Priest, J. G. Shapter, D. Losic, "*Influence of nanopore dimensions on electrochemical properties of nanopore arrays studied by impedance spectroscopy"* Sensors, 2014 (under review)
3. S. Chandrasekaran, M. J. Sweetman, **K. Kant**, W. Skinner, D. Losic, T. Nann and N. H. Voelcker, "*Silicon diatom frustules as nanostructured photoelectrodes*", Chemical Communications , (2014)
4. **K. Kant**, C. Priest, J. G. Shapter, D. Losic, "*Characterization of impedance biosensing performance of single and nanopore arrays of anodic porous alumina fabricated by focused ion beam (FIB) milling*" Electrochemica Acta 139 (2014) 225–231.
5. S. Simovic, K. R. Diener, A. Bachhuka, **K. Kant**, D. Losic, J. D. Hayball, M.P. Brownc, and K.Vasilev, *"Controlled release and bioactivity of the monoclonal antibody rituximab from a porous matrix: a potential in situ therapeutic device"* Materials Letters 130 (2014) 210–214.
6. **K. Kant**, J. Yu, C. Priest, J. G. Shapter and Dusan Losic *"Impedance nanopore biosensor: influence of pore dimensions on biosensing performance*" Analyst, (RSC), 139 (2014) 1134-1140.
7. **K. Kant,** M. Kurkuri, J. Yu, J. G. Shapter, C. Priest, D. Losic, *"Impedance spectroscopy study of nanopore arrays for biosensing applications"* Science of Advanced Materials, 6, (2014) 1375-1381.

8. Y. Yu, **K. Kant,** J. G. Shapter, J. Addai-Mensah, D. Losic, "*Characterisation of catalytic properties of gold nanotube membranes"*, Microporous and Mesoporous Materials, 153 (2012) 131-136.

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1. **K. Kant**, S. Amos, M. Erkelens, C. Priest, J. G. Shapter, D. Losic, '*Microchip nanofludic channel for microbial cell lysis and nucleic acids extraction'* RACI National Congress 7-12 Dec. 2014, Adelaide, Australia.
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