University of Victoria | Graduate Studies

Notice of the Final Oral Examination
for the Degree of Doctor of Philosophy

of

# AHMED AL BALUSHI

MSc (University of Bath, 2007)
BEng (University of Bath, 2005)

## "Double Nanohole Aperture Optical Tweezers: Towards Single Molecule Studies"

Department of Electrical and Computer Engineering

May 10, 2016
10:00 A.M.
Engineering Office Wing
Room 430

Supervisory Committee:
Dr. Reuven Gordon, Department of Electrical and Computer Engineering, University of Victoria (Supervisor)
Dr. Poman So, Department of Electrical and Computer Engineering, UVic (Member)
Dr. Stephanie Willerth, Department of Mechanical Engineering, UVic (Outside Member)

External Examiner:
Dr. Jer-Shing Huang, Department of Chemistry, National Tsing Hua University

Chair of Oral Examination:
Dr. Julian Lum, Department of Biochemistry and Microbiology, UVic

Dr. David Capson, Dean, Faculty of Graduate Studies

## Abstract

Nanoaperture optical tweezers are emerging as useful tools for the detection and identification of biological molecules and their interactions at the single molecule level. Nanoaperture optical tweezers provide a low-cost, scalable, straight-forward, high-speed platform for single molecule studies without the need to use tethers or labeling. This thesis gives a general description of conventional optical tweezers and how they are limited in terms of their capability to trapping biological molecules. It also looks at nanoaperture-based optical tweezers which have been suggested to overcome the limitations of conventional optical tweezers. The thesis then focuses on the double nanohole optical tweezer as a tool for trapping biological molecules and studying their behaviour and interactions with other molecules. The double nanohole aperture trap integrated with microfluidic channels have been used to detect single protein binding. In that experiment a double-syringe pump was used to deliver biotin-coated polystyrene particles to the double nanohole trapping site. Once stable trapped of biotin coated polystyrene particle was achieved, the double-syringe pump was used to flow in streptavidin solution to the trapping site. In addition, the double nanohole optical tweezer has been used to observe the real-time dynamic variation in protein-small molecule interaction (PSMI) with the primary focus on the effect of single and multiple binding events on the dynamics of the protein in the trap. In those studies, four forms of streptavidin were considered: bare streptavidin, bare monovalent streptavidin, biotinylated streptavidin and biotinylated monovalent streptavidin. Time traces of the trapping events of each one of the four forms were compared, with the bare form of the streptavidin showing slower timescale dynamics as compared to the biotinylated forms of the protein. Furthermore, the double nanohole aperture tweezer has been used to study the real-time binding kinetics of PSMIs and to determine their disassociation constants. The interaction of blood protein human serum albumin (HSA) with tolbutamide and phenytoin was considered in that study. The dissociation constants of the interaction of HSA with tolbutamide and phenytoin obtained using our technique were in good agreement with the values reported in the literature. These results would open up new windows for studying real-time binding kinetics of protein-small molecule interactions in a label-free, free-solution environment, which will be of interest to future studies including drug discovery.