The Final Oral Examination
for the Degree of

DOCTOR OF PHILOSOPHY
(Department of Physics and Astronomy)

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2008 University of Victoria MSc
2005 University of Victoria BSc (Honours)

“Investigating the use of protein-targeted pegylated gold
nanoparticle probes in the surface-enhanced Raman
spectroscopy of cells”

Thursday, December 18, 2014
1:00 PM
Elliott Building, room 105

Supervisory Committee:
Dr. Andrew Jirasek, Department of Physics and Astronomy, UVic
(Supervisor)
Dr. William Ansbacher, Department of Physics and Astronomy, UVic (Member)
Dr. Geoffrey Steeves, Department of Physics and Astronomy, UVic (Member)
Dr. Alexandre Brolo, Department of Chemistry, UVic (Outside Member)

External Examiner:
Dr. Michael Xiaoke Chen, Department of Physics
Simon Fraser University

Chair of Oral Examination:
Dr. Kieka Mynhardt
Department of Mathematics and Statistics, UVic
Abstract

Currently, it is very challenging to accurately monitor the response of patients to radiation therapy over the course of treatment. The initial response to ionizing radiation occurs in the cells at a molecular level, and effects of the response are not typically noticeable on short time scales. Surface-enhanced Raman Spectroscopy, or SERS, has proven to be a useful technique in the analysis of tissues and cells at a molecular level. Specifically, the use of targeted SERS probes allows for the detection of specific proteins on the cell membrane. The work presented here looks to assess the feasibility of using targeted SERS probes and two-dimensional SERS microscopy to measure the response of tumour cells to ionizing radiation, by identifying changes in the distribution of membrane proteins following exposure to clinically relevant doses of ionizing radiation (≤ 60Gy).

Two different types of targeted SERS probes were investigated, based on the work of Grubisha et al. ([1]; Type I) and Qian et al. ([2]; Type II), both containing a gold nanoparticle core. In a simplified cellular experiment, biotin on the surface of biotinylated OVCAR5 cells was targeted with streptavidin-SERS probes, and the Type-II SERS probes showed the most promising results. However, SERS maps still provided less characteristic spectral signal than expected, and challenges remain in the development of a reproducible cellular imaging technique.

Despite difficulties in cellular imaging, the functionality of the Type-II SERS probes was verified separately, using gold slides with a biotin monolayer in place of cells. Following verification, the SERS intensities provided by differently sized clusters of the SERS probes were characterized. To begin, both SERS maps and scanning electron microscope (SEM) images of gold slides were acquired after incubation with Type-II SERS probes for multiple times (1hr, 2hr, 3hr, 12hr). Data analysis of the SEM images provided a measure of the physical distribution of the SERS probes on the surface of the slide, while analysis of the SERS maps provided information about the spectral distribution of the probes. By relating the information provided by the SEM images and SERS maps, a simple polynomial relationship between SERS intensity and the number of clustered SERS probes providing the enhancement was determined, providing a framework for quantifiable SERS imaging.

Finally, an independent experiment was devised to ensure that exposure to clinically relevant doses of ionizing radiation would affect the ability of the targeted protein to bind to SERS probes, thus leading to measurable differences in SERS maps of irradiated and unirradiated cells. A series of
experiments utilizing the enzyme-linked immunosorbant assay (ELISA) was performed to test the effect of ionizing radiation-induced damage on the ability of streptavidin to bind to biotin, and the results confirmed that a noticeable reduction in binding could be detected at doses as low as 10 Gy.

The results of this work demonstrate that following the development of a suitable cell/SERS probe incubation technique, Type-II SERS probes would be appropriate for use in quantifiable SERS imaging. Also, it is suggested that a measurable change in protein function will be present when comparing SERS maps of control cells to those of cells irradiated to clinically relevant doses.


Awards, Scholarships, Fellowships

2008-2012 – Canada Graduate Scholarship, PGS-D, NSERC
2008-2012 – President’s Research Scholarship, University of Victoria
2007 – Nora and Mark Degoutiere Memorial Scholarship, University of Victoria
2005-2007 – University of Victoria Fellowship, University of Victoria

Presentations


**Publications**


