ND*nano* Undergraduate Research Fellowship (NURF)

1. Student name: Colleen Riordan

2. Faculty mentor name: Zachary Schultz

3. Project title: SERS Detection of Glucose Phosphate Isomers

4. Briefly describe any new skills you acquired during your summer research:

This summer I learned to do kinetic studies in order to determine how long it takes for the glucose phosphates to bind to the SERS substrate by analyzing the intensity of the glucose phosphate peaks over time. I also learned how to operate the flow cell and CE developed by the Schultz lab.

5. Briefly share a practical application/end use of your research:

Glucose phosphates are important in metabolism since glucose 6-phosphate is an essential step in the breakdown of glucose to pyruvate for energy while glucose 1-phosphate is an essential step in the storage of glucose as glycogen. An ability to track the ratios of glucose 1-phosphate to glucose 6-phosphate under different circumstances could provide valuable information to metabolomics- identifying changes in biological pathways relevant to health and disease.

Project Summary:

The identification and separation of isomeric carbohydrates is notoriously difficult. As the specific structure of carbohydrates correlate to distinct functions, it is essential to be able to distinguish between different isomeric structures. The most common methods for analyzing carbohydrates are nuclear magnetic resonance (NMR) and mass spectrometry (MS), but innate weaknesses of both methods mean that neither option is perfectly suited for distinguishing carbohydrates. An alternative approach to MS and NMR is Raman spectroscopy. In Raman spectroscopy monochromatic light is irradiated on the analyte of interest causing the photons to be raised from ground state to some virtual state and then scattered at different wavelengths that correspond to the sample's vibrational frequencies. These vibrational frequencies are unique to each structure, allowing for the differentiation of isomeric analytes. One disadvantage of Raman is low sensitivity, since only $1x10^{-13}$ photons are scattered meaning small sample sizes are hard to detect, leading to the development of SERS. Surface-enhanced Raman spectroscopy (SERS) surfaces are typically composed of silver or gold nanoparticles, because these metals have a localized plasmon resonance which creates an electric field when irradiated with the laser. (Yoshida et al) The interaction of the electric field with the electrons of the analyte enhances the scattered photons collected by many orders of magnitude (Moskovits) which enables detection of analyte at low concentrations. The biggest disadvantage of SERS is that in order to achieve this enhancement, the analyte must be extremely close to the SERS substrate to interact with the electric field. When attempting to collect SERS spectra of an analyte in solution, getting the analyte close enough to the SERS surface to achieve the enhanced signal can be difficult. The goal of my project is to use Raman, and specifically SERS, to identify the differences in the

spectra of simple isomeric carbohydrates. This summer I worked on developing a technique for detecting and separating isomers of glucose phosphate at biologically relevant concentrations.

One solution to the issue of analyte proximity to the surface is to use a self-assembled monolayer (SAM) on the SERS surface. The basic idea is that the SAM interacts with the analyte of interest, which brings the analyte into close enough proximity to the surface that the Raman enhancement is achieved. Most SAM's that are applied to SERS substrates have a thiol group at one end, as thiols have a high affinity for noble metals, and a group at the other end that can interact with the analyte of interest. (Love et al) Preliminary results I have obtained show that a SAM of 4-mercaptophenylboronic acid (4-MPBA) or 3-MPBA are both able to bind glucose 6-phosphate and show a marked change in SERS spectra (Figure 1) after the substrate was left soaking in a 1 mM glucose 6-phosphate solution overnight. I also attempted kinetic studies, in which I used a water-immersion lens on the Raman microscope to track the binding of glucose phosphates to the surface in real-time. Though there were some promising results in the kinetic studies (Figure 2) overall my results failed to show reproducibility and appeared to have high amounts of dirt contamination, both of which are often problems encountered with SERS.

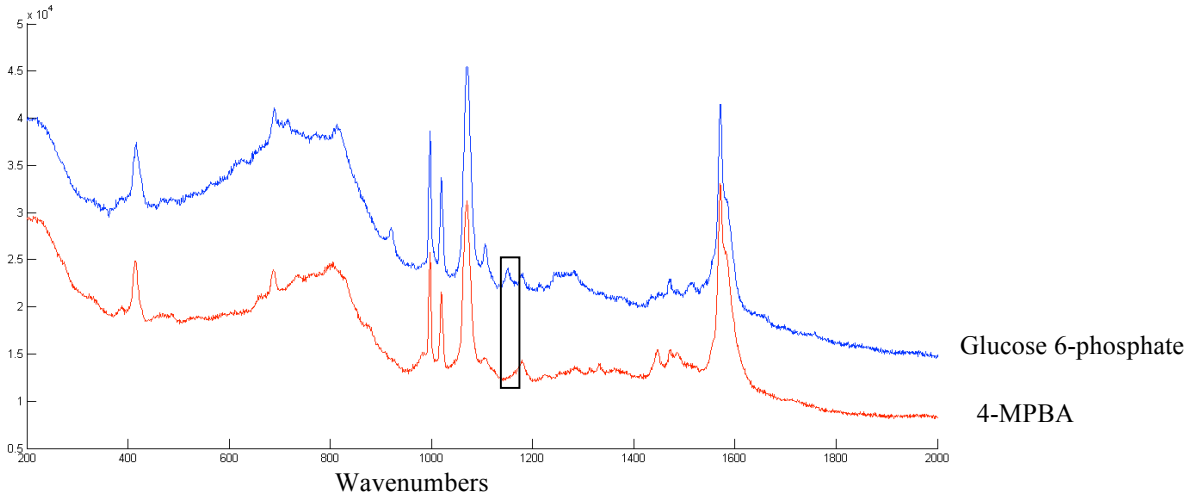

Figure 1: SERS spectra of glucose 6-phosphate as compared to the 4-MPBA background.

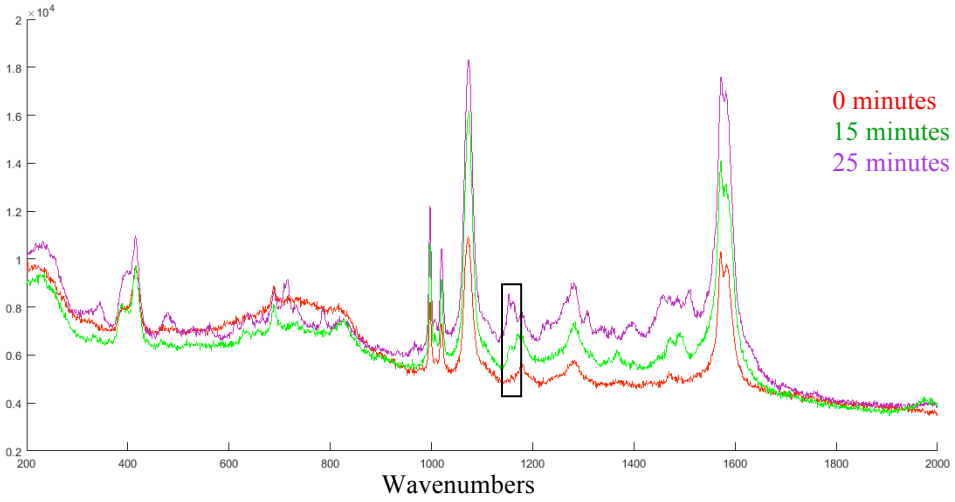

Figure 2: Kinetic SERS study of glucose 6-phosphate on 4-MPBA SAM with 1150 cm^{-1} peak growing in over time

References:

Love, J; Estroff, L; Kriebel, J; Nuzzo, R; Whitesides, G. Self-Assembled Monolayers of Thiolates on Metals as a Form of Nanotechnology. *Chem. Rev.* ***2005***, *105*, 1103−1169.

Moskovits, M. Surface-Enhanced Raman Spectroscopy: A Brief Retrospective. *Journal of Raman Spectroscopy* ***2005***, *36*, 485-496.

Yoshida, K.; Itoh, T.; Tamaru, H.; Biju, V.; Ishikawa, M.; Ozaki, Y. Quantitative Evaluation of Electromagnetic Enhancement in Surface-Enhanced Resonance Raman Scattering from Plasmonic Properties and Morphologies of Individual Ag Nanostructures. *Phys Rev B* ***2010***, *81*, 9.

Publications (papers/posters/presentations):

- Abstract accepted for poster presentation at SciX, a national meeting for the Society of Applied Spectroscopy; October 2015
- Poster presentation at Notre Dame Undergraduate Research Symposium; July 2015