Detection of 3,3’-dihexyloxacarbocyanine iodide by SERS inside the silica/silver colloid sol-gel matrix

Sean Webster; Dr. Elizabeth J. O. Atkinson

Department of Chemistry, Linfield University, McMinnville, Oregon 97128

Introduction

The term sol-gel can be derived from a process in which the hydrolysis of precursor molecules in solution agglomerate to form a three-dimensional coherent network. In this investigation, SiO_2 was used as the precursor molecule because of its ability to form porous, highly structured matrices. In recent years, silica sol-gels (and sol-gels in general) have drawn increasing interest for their applications as biosensors and because of their high emissivity coating: making it an energy-saving material used commonly in spacecraft and industrial furnaces[1]. It should also be noted that sol-gels are the precursor to aerogels, with the difference being that a sol-gel contains a solvent trapped inside its matrix, whereas an aerogel has had its solvent removed. Furthermore, silver colloid-modified SiO_2 sol-gels make-up a unique class of materials that can serve as substrates for Surface-Enhanced Raman Spectroscopy (SERS) measurements.

Moreover, SERS is a spectroscopic technique that is caused by the inelastic scattering of photons from molecules and can be used to identify single target molecules that are bound to noble metal nanoparticles under the proper conditions. This technique is useful because a target molecule produces a unique spectrum distinguishable from other compounds, allowing for accurate identification.

In this research study, an organic fluorescent charged dye, 3,3’-dihexyloxacarbocyanine iodide $DiOC_6(3)$ (see Figure 1 for chemical structure), was chosen as a target molecule for detecting distinguishable SERS signals and as a basis for Ultraviolet-Visible (UV/Vis) Spectroscopy in base-catalyzed and acid-catalyzed silver colloid sol-gels because of its facilitating ability to stain mitochondria and identify mitochondrial changes during early apoptosis in animal and plant cells[2], and for its capability to stain and visualize thrombus formation and platelet aggregate in red blood samples[3]. Thus, the detection of $DiOC_6(3)$ within modified sol-gels could lead to the development of new techniques for staining and characterizing other biological, physical, and electrochemical matter as SERS-based sensors.

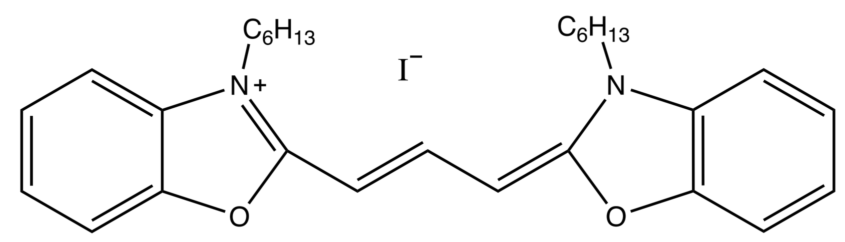

Figure 1: Chemical structure of 3,3’-dihexyloxacarbocyanine iodide.

Materials and Methods

The following chemical was purchased through Quartzy (Hayward, CA): $DiOC_6(3)$.

Ag Colloid preparation:
All glassware utilized was thoroughly cleaned with Aqua Regia (3:1, conc. HCl:conc. HNO_3), rinsed with tap water, then rinsed and soaked in DI H_2O to remove any organic material. $AgNO_3$ (100 mL, 1 mM) and DI H_2O (50 mL) were brought to a boil with stirring. Sodium citrate (14.0 mL, 1% by weight) was slowly added (1 drop/10 seconds) and the solution was boiled for an additional 30 minutes. The resulting solution was allowed to cool to room temperature (~25 ºC) and was stored in the refrigerator.

Base-catalyzed sol-gel synthesis:
Methanol (7.38 mL), tetramethyl orthosilicate (3.47 mL), DI H_2O (0.373 mL), $DiOC_6(3)$ (1.00 mL, 0.00050 M), and NH_4OH (0.0198 mL, 30%) were combined and stirred for five minutes. Ag Colloid (2.00 mL) and Na_2SO_4 (1.00 mL, 1.0 M) were added to the mix, and the solution was stirred for an additional minute. The resulting solution was poured into plastic cuvettes to allow for gelation and aged for approximately 24 hours.

Acid-catalyzed sol-gel synthesis:
In the following order, DI H_2O (6.62 mL), HCl (0.0036 mL, 0.04 M), tetramethyl orthosilicate (4.62 mL), and $DiOC_6(3)$ (1.00 mL, 0.00021 M) were combined and stirred for fifteen minutes. Ag Colloid (2.00 mL) and Na_2SO_4 (1.00 mL, 1.0 M) were added to the mix, and the solution was stirred for an additional minute. The resulting solution was poured into plastic cuvettes to allow for gelation and aged for approximately 72 hours.

Surface-Enhanced Raman Spectroscopy (SERS):
SERS spectra were acquired using a custom-built Raman spectrometer with a 532 nm laser excitation and a 50 nm slit size.

Results

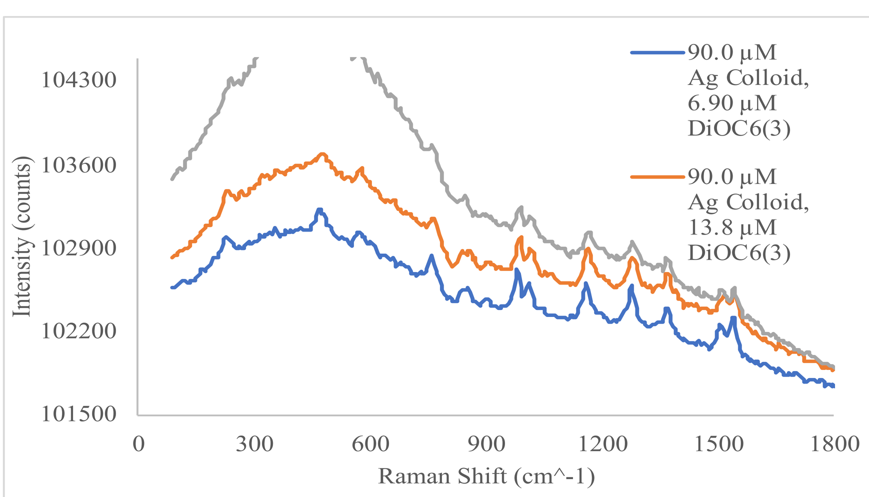

Figure 2: SERS spectra of acid-catalyzed sol-gels with a constant silver nanoparticle concentration and varying $DiOC_6(3)$ concentrations, taken immediately after gelation.

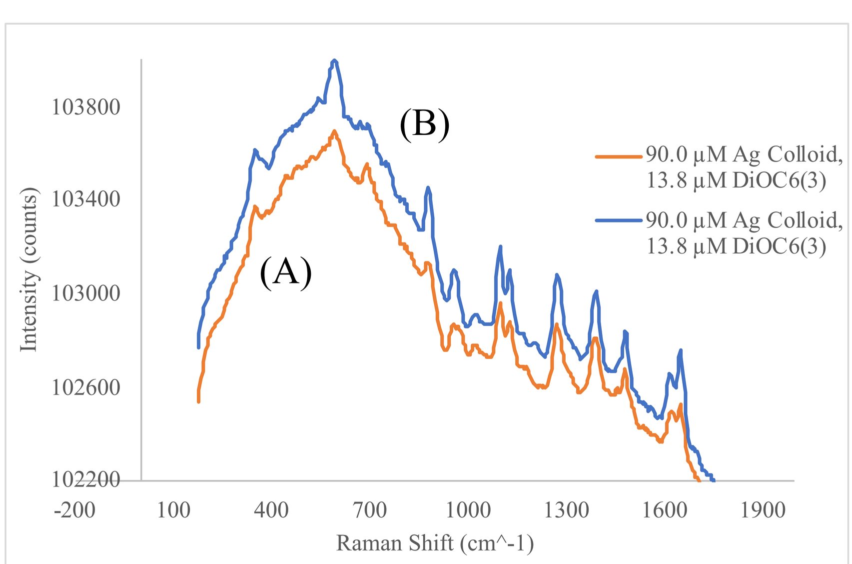

Figure 3: SERS spectra of acid-catalyzed sol-gels containing silver nanoparticles and $DiOC_6(3)$, taken immediately after gelation (A) and 24 hours after gelation (B).

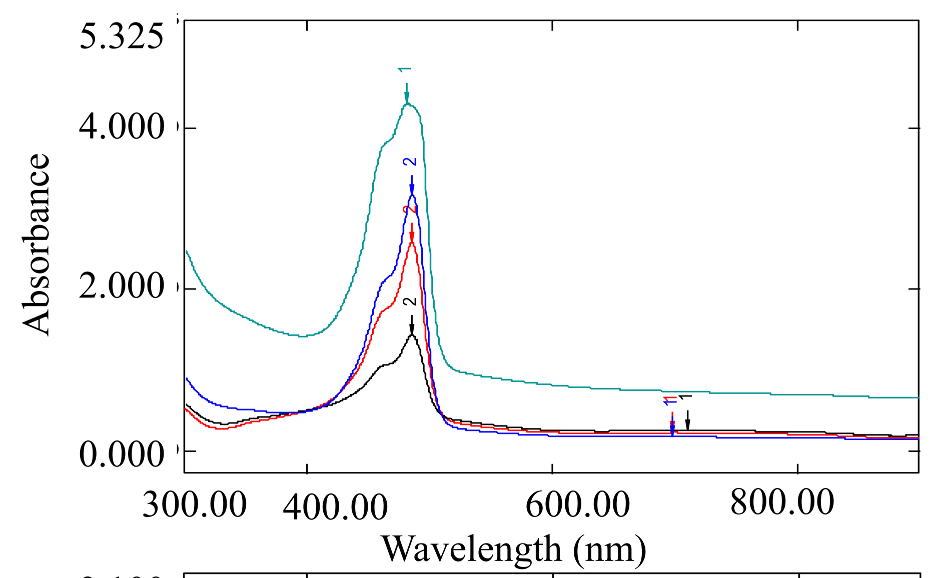

Figure 4: UV/Vis spectra of acid-catalyzed sol-gels with a constant silver nanoparticle concentration and varying $DiOC_6(3)$ concentrations, taken immediately after gelation.

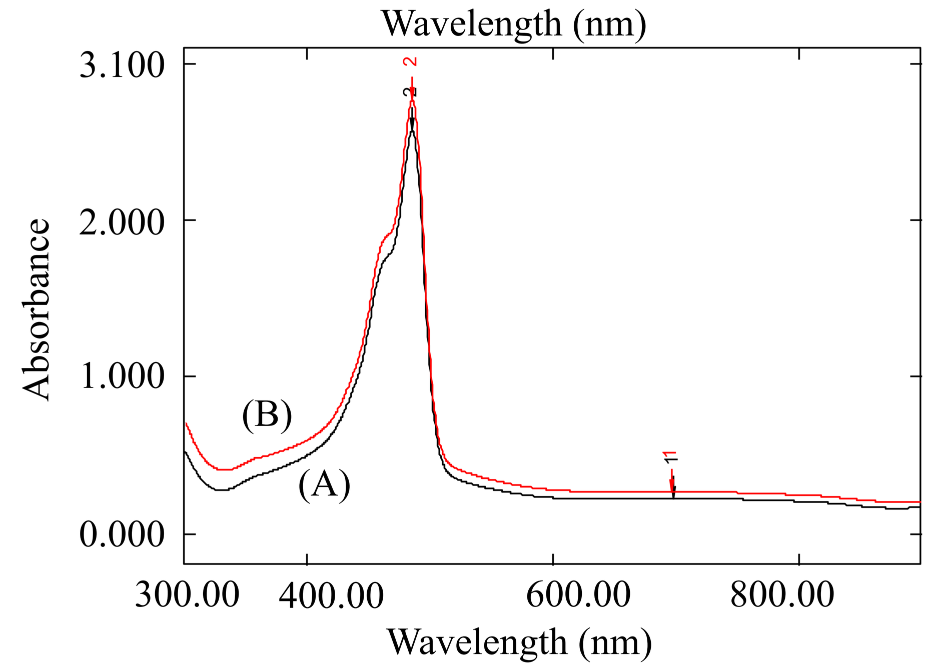

Figure 5: UV/Vis spectra of acid-catalyzed sol-gels containing silver nanoparticles and $DiOC_6(3)$, taken immediately after gelation (A) and 24 hours after gelation (B).

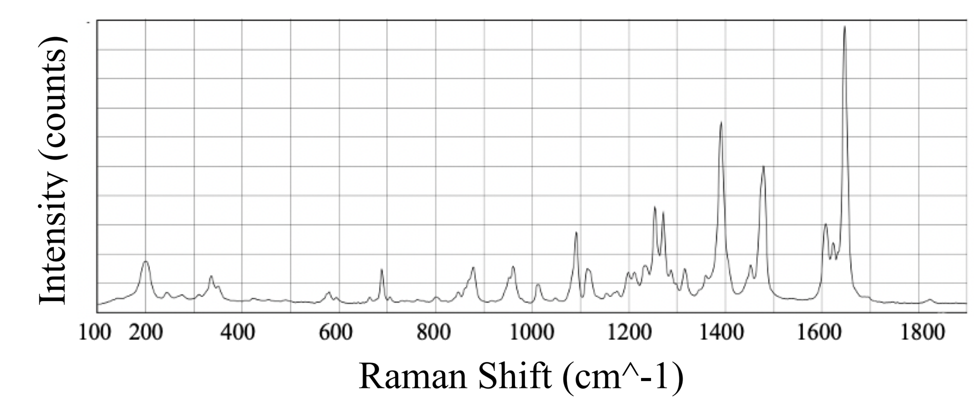

Figure 6: Published Raman spectrum of $DiOC_6(3)$ from Sigma Aldrich.[4]

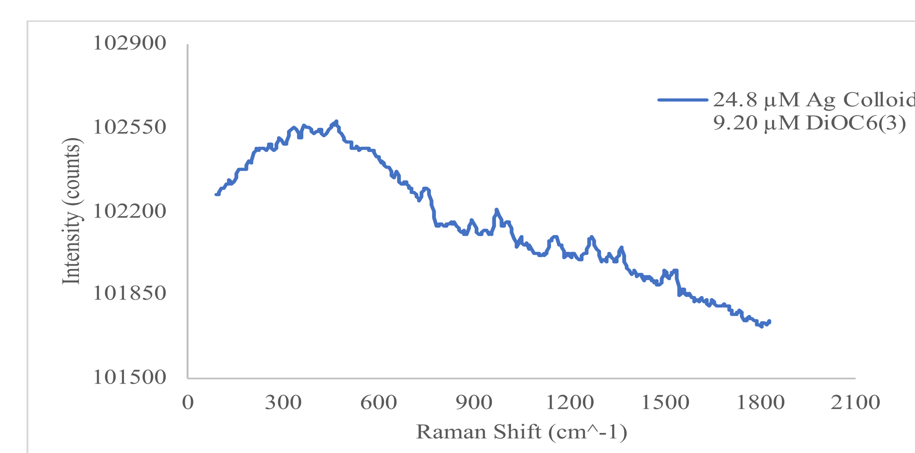

Figure 7: SERS spectrum of a base-catalyzed sol-gel with silver colloid and $DiOC_6(3)$, taken immediately after gelation.

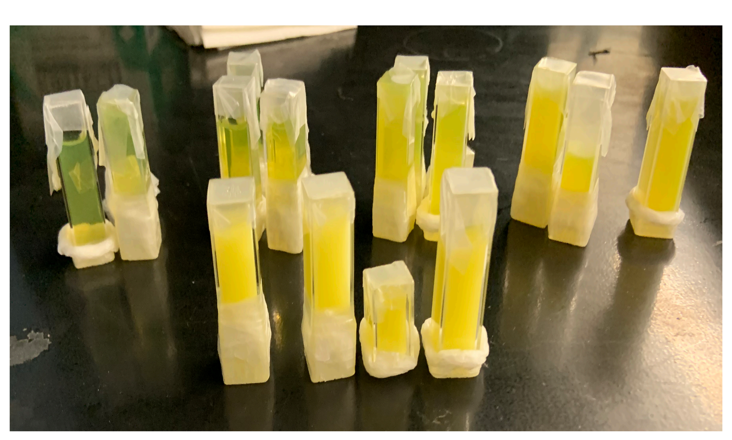

Figure 8: Image taken of acid-catalyzed sol-gels in plastic cuvettes after 24 hours of aging containing silver colloid and $DiOC_6(3)$. Concentration increases going from left to right.

Conclusion

The results from Figures 2 and 3 suggest that SERS spectra of DiOC6(3) were experimentally obtainable and that there was a high degree of stability present for DiOC6(3) within acid-catalyzed silica sol-gels (see Figure 6 for the published Raman spectra of DiOC6(3)). This high level of stability was correlated with the stability of silver nanoparticles as shown in Figures 4 and 5 through Ultraviolet-visible (UV/Vis) spectrophotometry. Thus, it is likely that the observed DiOC6(3) SERS signal stability of the positively charged dye led to the stabilization of the silver nanoparticles within the silica sol-gel matrix.

Contrarily, it should be noted that although a SERS signal for DiOC6(3) within a base-catalyzed silica sol-gel was experimentally obtainable immediately after gelation (see Figure 7), but the signal was lost after 24 hours. This is likely because DiOC6(3) is a charged species, thus, interacting with other species in solution in a way that causes the signal to decay.

From these findings, it was ultimately concluded that acid-catalyzed sol-gels containing silver nanoparticles are viable substrates for SERS of DiOC6(3) and that these materials could lead to the development of new techniques for detecting mitochondrial DNA and characterizing other biological, physical, and electrochemical matter as SERS-based sensors.

Literature Cited

[1] Mahadik, D., Gujjar, S., Gouda, G. and Barshilia, H., 2020. *Double Layer Sio2/Al2o3 High Emissivity Coatings On Stainless Steel Substrates Using Simple Spray Deposition System*. [online] ScienceDirect. Available at: <https://www.sciencedirect.com/science/article/abs/pii/S0169433214002116> [Accessed 27 September 2020].

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Future Studies

Future investigations will take particular interest in observing the stabilization of $DiOC_6(3)$ in acid-catalyzed aerogels and xerogels, integrating polystyrene beads inside the sol-gel/aerogel matrix, and obtaining Atomic Force Microscopy (AFM) images of base-catalyzed/acid-catalyzed sol-gels.

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Further Information

Please contact Elizabeth Atkinson at eatkins@linfield.edu for any further information.