

Synthesis and Characterization of Coated Gold Nanoparticles with Embedded SERS Tags



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Introduction

Background: Nanoparticles are finding widespread use in many fields such as healthcare and the environment.^{1,2} However, they are of particular importance as drug delivery vehicles in biological systems. By tagging them with therapeutic drugs or antibodies and coating them in a phospholipid bilayer they have been found to be biocompatible and enter cells².

Surface-enhanced Raman spectroscopy (SERS) is of particular importance as an optical bioimaging technique due to its ability to allow deep and high-resolution volumetric imaging of biological tissues. Moreover, SERS can even allow for single molecule detection¹. For a drug delivery construct to be monitored in-vivo, a SERS active molecule must be adsorbed close to or on the surface of metal nanoparticles. By using a 532 nm laser, the plasmon resonance of silver is more intense than gold, therefore silver was coated on the Au-NP.

Goals: In this study, gold nanoparticles (Au-NPs) have been synthesized using a modified seed-mediated method,^{1,3} coated with *para*-mercaptobenzoic acid (pMBA) (Figure 1), a SERS active molecule, followed by the addition of silver to allow detection from the Raman spectrophotometer to provide greater SERS enhancement, and finally a phospholipid bilayer to promote uptake of the particles into biological systems.

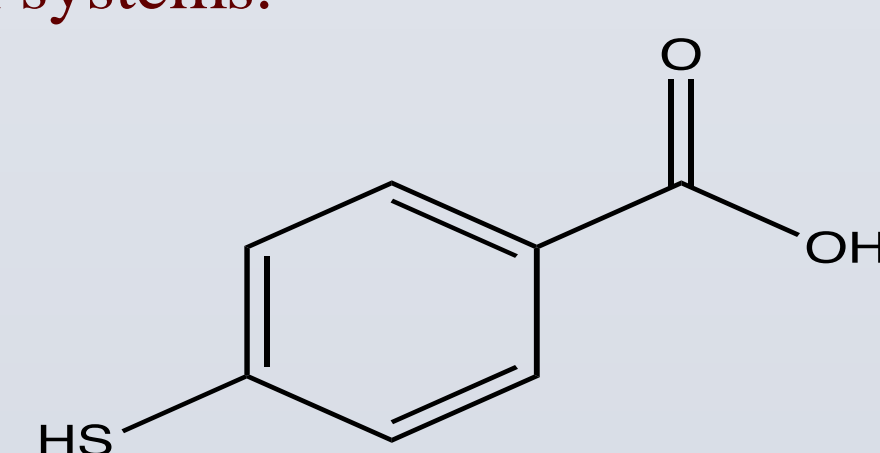


Figure 1. *para*-mercaptobenzoic acid (pMBA)

Materials and Methods

Chemicals: Gold(III) chloride hydrate, sodium citrate tribasic hydrate, 4-mercaptobenzoic acid (pMBA), silver nitrate, cetyltrimethylammonium chloride (CTAC), ascorbic acid, and poly(allylamine hydrochloride) (PAH) were purchased from Sigma-Aldrich (St. Louis, MO). 1-Palmitoyl-2-oleoyl-*sn*-glycero-3-phospho-L-serine (POPS), and lysophosphatidylcholine (LPC) were purchased from AvantiPolar Lipids(Alabaster, Alabama). All the solvents and reagents were analytical grade.

Au-NP synthesis: All glassware was cleaned with Aqua Regia (3:1, conc. HCl:conc. HNO₃) then rinsed with deionized water. Chloroauric acid (3.0 mL, 0.01 M) and deionized water (88 mL) were refluxed until boiling. Sodium citrate (1%, 5.0 mL) was added and the solution was refluxed for an additional 30 minutes until the solution turned a light red color and the solution was cooled on ice.

Silver coating: Au-NP (3 mL) and pMBA (15 μ L, 2.5 mM, ethanol) were sonicated and silver chloride (1.2 mL, 1 mM), CTAC (3 mL, 0.1 M) and ascorbic acid (150 μ L, 1 M) were added to the Au-NP and pMBA solution.

PAH and lipid coating: PAH (200 μ L, 10 mg/mL) and sodium chloride (100 μ L, 0.1 M) were immediately added to 1 mL of the solution and purified. POPS/LPC lipid solution (0.5 mL, 1:1 w/w) was added to the solution, incubated, and purified.

Spectroscopy analysis: Au-NP were characterized using a Hitachi UV-vis spectrometer, NanoSight LM10HS particle size analyzer, and the extinction spectra were recorded. Spectra of coated BRIGHTs containing pMBA were obtained with a custom-built Raman spectrometer using a 532 nm laser and 50 μ m slit width. Data was acquired using KestrelSpec at 10 second acquisition times with an automatic background subtraction.⁵

Results

Table 1. Approximate diameter of Au-NP at various stages of synthesis.

Gold Nanoparticle Solution	Diameter (nm)	Gold Nanoparticle Solution	Diameter (nm)
Stock Solution Au-NP	16	With 0.30 mM Ag shell	60
With pMBA pre-purified	29	With PAH post-purification	75
With pMBA post-purified	81	With Lipids post-purification	125
With 0.28 mM Ag shell	55		

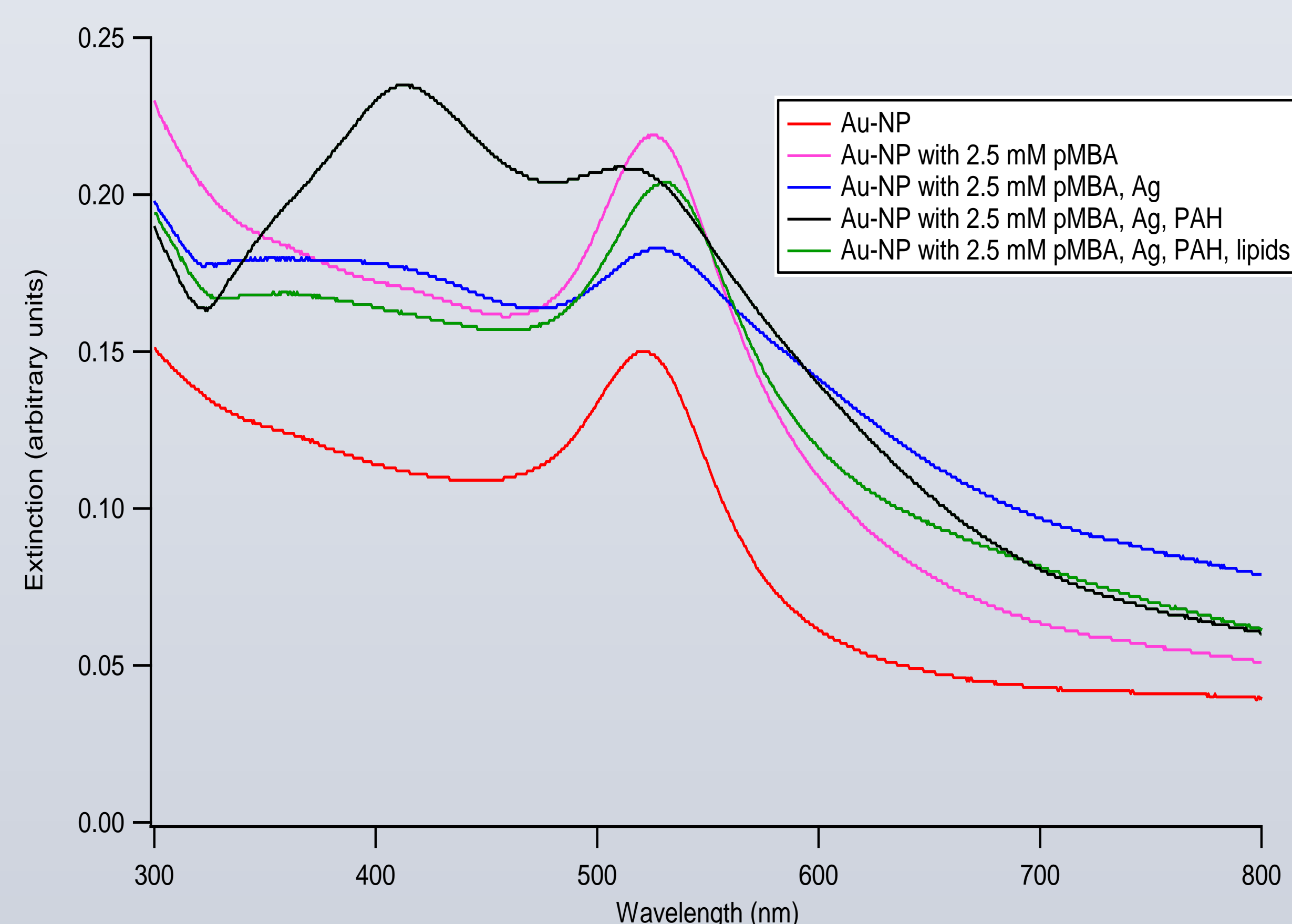


Figure 1. UV-vis extinction spectra of Au-NP with 2.5 mM pMBA, 0.28 mM Ag, PAH and lipid coatings.

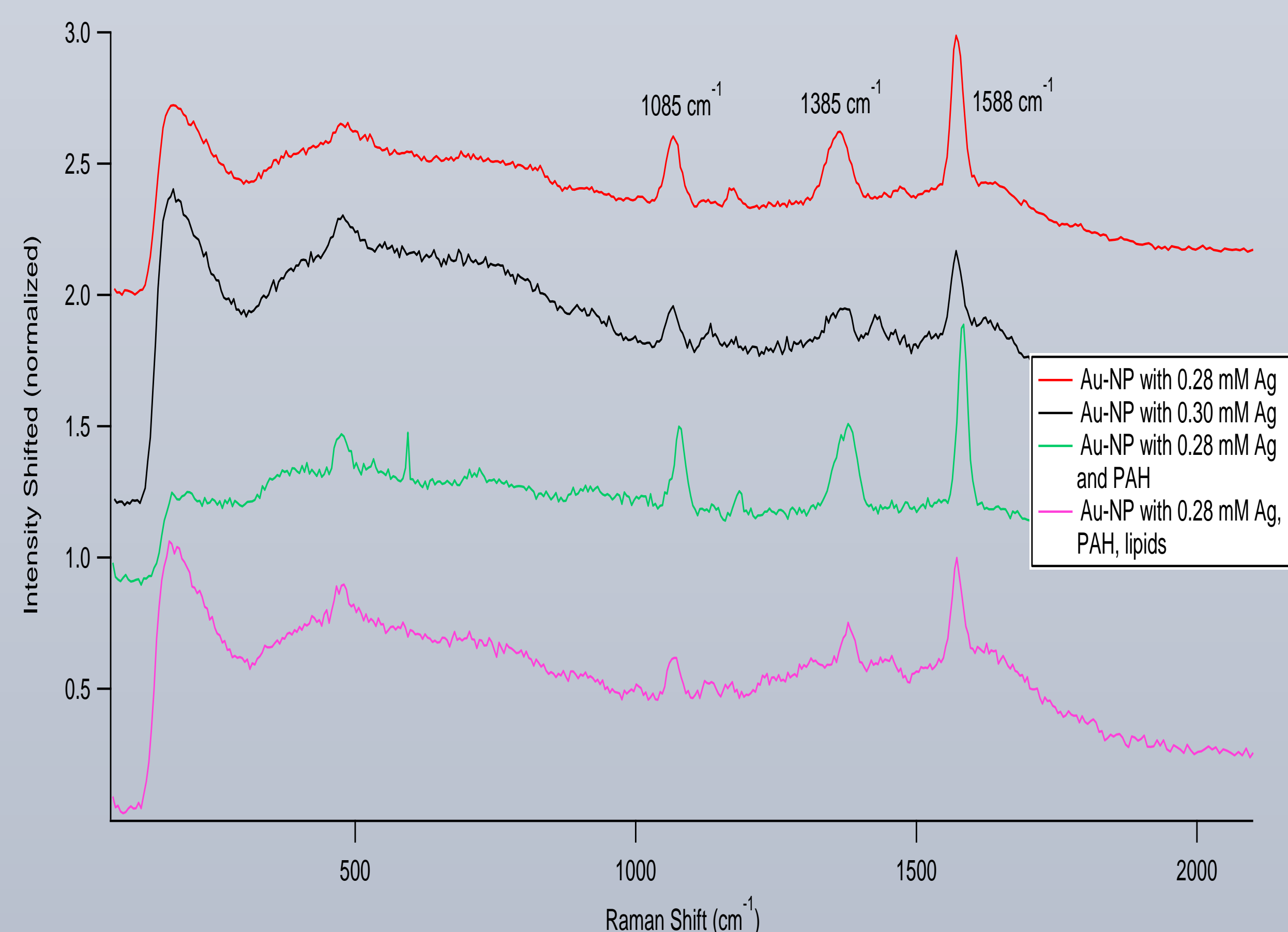


Figure 2. Offset of normalized SERS spectra of Au-NP with 2.5 mM pMBA, Ag (0.28 and 0.30 mM), PAH and lipid coatings.

Discussion

The diameter of the Au-NPs increased with each additional coating (Table 1). Small increases in diameter indicated successful coating of Au-NP. Silver coated BRIGHT's greatly increased in size when purifying directly after silver coating indicating aggregation. This aggregation was reduced by immediately coating with PAH after silver.

Figure 1 shows UV-vis spectra of Au-NPs throughout the coating process. Initial Au-NPs showed a single peak at 521.50 nm and experienced a red shift with each additional coating of the particles. Two peaks are observed in the spectra of particles containing silver indicating that the silver successfully coated the Au-NPs.

Figure 2 shows SERS spectra of pMBA throughout the coating process once the silver shell was synthesized. Characteristic peaks of pMBA can be seen at 1588 and 1085 cm^{-1} signaling aromatic ring vibrations.⁴ Another distinct peak can be seen at 1385 cm^{-1} corresponding to the carboxylate group of pMBA.⁴

Conclusions

Small increases in diameter, red shifts in the UV-vis spectra and the constant characteristic SERS peaks of pMBA suggest that coating of the Au-NP with pMBA, Ag, PAH, and lipids was successful while still being able to observe the SERS spectra of pMBA.

Further research will include:

- Increasing the stability of BRIGHTs by altering the concentrations of silver and PAH
- Monitoring cellular uptake in yeast cells
- Coating Au-NP with antibodies to control uptake into biological systems and target specific tissues

References

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Acknowledgements

We would like to thank the Linfield College Student-Faculty Collaborative Research Grant for funding this Research. We would also like to thank the Linfield Chemistry department, especially Dr. Thomas Reinert, Dr. Megan Bestwick, and Dr. Elizabeth Atkinson for assistance throughout the research project. An additional thank you to fellow research partners Chris Munjar, Madison Gladding, and Victoria Wood.