Raman-active gold nanoparticles as beacons in cervical cancer cells

Jennifer Monahan1, Tatjana Chernenko1,2, Amit Singh2, Miloš Miljković1, Mansoor Amiji3, Max Diem1

1Department of chemistry and Chemical Biology, Northeastern University, 360 Huntington Avenue, Boston, MA, USA 02115
2Department of Pharmaceutical Sciences, Northeastern University, 360 Huntington Avenue, Boston, MA, USA, 02115

Introduction

In 2010, it was estimated that over 1.5 million people would be diagnosed with cancer in the United States. Unfortunately, the existing forms of chemotherapeutic treatments often fail in delivering the drug to the tumor site, resulting high mortalities and drug resistance. Thus, the ability to reliably deliver drugs through a nanoparticle-based system is of great interest.

Nanotechnology improves accuracy, efficacy, speed, and safety in medicine. Specifically, engineered nanoparticles allow for biocompatibility by various surface modifications and targeting of biochemical components within the body. This allows the patient to no longer be exposed to lethal doses of chemotherapy and directly delivers the drug to the tumor site.

We proposed that a nanoparticle system composed of a spherical gold core modified with cysteine coupled to 2-cyano hexenoic acid, 2-cyano hexenoic acid acted as a surface-enhanced Raman spectroscopy (SERS) reporter molecule in which the cyano vibration occurred in a region devoid of any cellular information (ca. 2200 cm^-1). The gold surface will eventually be modified with polyethylene glycol to aid in biocompatibility along with a targeting moiety.

The nanoparticle system was characterized and incubated with HeLa cells at various concentrations and times. The induced biochemical changes were monitored via Raman micro-spectroscopy in which the SERS reporter acted as a tracking beacon upon nanoparticle internalization. Multivariate analysis techniques, namely Vertex Component Analysis (VCA), was utilized to understand and image the cellular responses to the nanoparticle system.

Results and Discussion

Nanoparticle characterization:

Raman spectra of 2-cyano hexenoic acid (A), Au-cysteine nanoparticles (B), and Au-cysteine-cyano nanoparticles (C). The Raman spectrum of 2-cyano hexenoic acid clearly shows the cyano vibration (ca. 2230 cm^-1) which was used to track the nanoparticles throughout the cell. As expected, the cyano vibration occurs in the Au-cysteine-cyano nanoparticles and not in the Au-cysteine nanoparticles.

Identification of nanoparticles within a cell:

The Raman spectrum below clearly indicates the cyano vibration (ca. 2230 cm^-1) due to the cellular architecture upon nanoparticle internalization. The 20X magnification of the cell is shown below and the red circle indicates where the inclusion was located along with the corresponding spectrum.

Variations in the cyano vibration:

Peaks due to the cyano vibration were seen at various points within a cell and shifting of its position was observed. This could be indicative of the subcellular environments and how the nanoparticles were sequestered within the cell.

Vertex Component Analysis:

The cell to the left was analyzed using VCA to identify the subcellular changes induced by nanoparticle internalization, and a pseudo color image was created to visualize these changes. Due to the burning and the high S/N, VCA analysis could only be performed on the cyano (ca. 2230 cm^-1) and the C-H stretching (ca. 2940 cm^-1) spectral regions. These two small spectral regions still provided enough information to locate the nucleus/nucleoli (blue), cytoplasm (green) and a small nanoparticle inclusion (red).

Conclusions

- The Au-cysteine-cyano nanoparticle design does exhibit an enhanced Raman spectrum and the cyano vibration is easily distinguished allowing for tracking within a cell.
- Upon incubation of the Au-cysteine-cyano nanoparticles in HeLa cells, the peak corresponding to the cyano vibration was found in various cytoplasmic and perinuclear regions.
- It is speculated that the observed shifting of the cyano peak may be due to how the cell sequesters the nanoparticles.
- Vertex component analysis was able to distinguish a nanoparticle inclusion near the peripheral of the nuclear membrane. However, because of the large concentration of nanoparticles, various areas within the cell needed to be excluded due to burning of the cell. It is hypothesized that the burned regions of the cells, correspond to very large nanoparticle inclusions.

References:

Acknowledgments:
Partial support from the ISERP Nanoscience and Technology Award DMR 0958342 (U.S.) and grant CA133159 (M.A.) from the US National Cancer Institute at the National Institutes of Health.