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Nanoparticles for Detection, Diagnostics, and Targeting using Hyperspectral Imaging

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Novel and modified nanoparticles containing multiple inherent and specific functionalities make them powerful tools in bioimaging, cancer targeting, cancer therapy, and microbial capture and detection.[1] For example, gold and iron oxide nanoparticles offer potential advantages in the bioimaging of cancer cells due to their respective strong absorption and scattering, and magnetic properties.[2] Several studies in the literature suggest the potency and efficiency of these nanoparticles in therapeutic applications. For example, per Alhalili Z. et al (2016), there was no observed significant decrease ($P>0.05$) in cell viability when TD47 cells are treated with gold nanoparticles (AuNPs) alone.[3] However, the same cells were significantly killed by the application of AuNPs chemically conjugated to Taxol, suggesting the potential of gold nanoparticles for efficient delivery of Taxol to breast cancer cells. Similarly, Ding R.L. (2017) reported the release of endostatin (ES) in a sustained manner *in vitro* that showed an excellent inhibitory effect on HUVECs proliferation and migration.[4] The study concluded that ES-NPs significantly improved the anticancer activity of ES by affecting angiogenesis. Despite similar developments in nanomaterial research, much remains to be done in terms of the characterization of the interaction between the cells and the drug loaded nanoparticles. In this study, we report the use of CytoViva Hyperspectral Imaging technology to identify cellular uptake of nanoparticles and to attempt to characterize (if any), the interaction between selected NPs and colon cancer cells.

The CytoViva's Hyperspectral microscopy technology incorporates patented high signal-to-noise optical microscopy with high spectral resolution hyperspectral imaging (HSI). This enables optical observation and quantitative spectral analysis of nanoscale samples in a wide range of biological and materials-based environments. In addition, the hyperspectral microscopy can be used to confirm unique surface chemistry and functional groups added to nanomaterials. Furthermore, certain biologicals, such as bacteria and pathogens, can be optically observed, spectrally characterized and mapped in tissue and other environments, especially after capturing or marking them with nanoparticles. Importantly, the imaging system does not require any fluorescent markers.

In this study, SW480 colon cancer cells (ATCC) were grown in chamber slides and exposed to varying concentrations (10-100 µM) of nanoparticles solutions in different wells. After an overnight exposure, the monolayers were fixed in 4% formalin in PBS and imaged using the CytoViva HSI system. In this preliminary work, we demonstrate the use of HSI to characterize the spectral profile of nanoparticles internalized by or attached to cells. Figures 1A and 1B show an optical image and hyperspectral scan of the SW480 cells, respectively, with no added nanoparticles. Figure 2A depicts the AuNPs inside cells or in circular rings around the cells suggesting surface attachment. The HSI spectra (Figure 2B) show very intense and sharp peaks with maxima at about 650 nm and intensity in the range of 4500-6000 counts. Note that even though the HSI technique doesn't give morphological details, it identifies specific particles based on their wavelength of absorption and spectral characteristics. Figures 3A and 3B are an HSI scan of bacteria with an overlay of spectral profiles from an independent specimen. This preliminary study indicates that differences in Hyperspectral profiles offer powerful tools that enable the determination of particle conjugation and their association with cells/bacteria. Further studies to enable and optimize intracellular detection are underway.

References

1. Kang X. et al. *Scientific Reports 7*, (2017) Article number: 42069 doi:10.1038/srep42069
2. R. Bardhan, l. S. La, A. Joshi, & N. J. Halas, Acc *Chem Res*. (2011) **44,** 936.
3. Z. Alhalili, D. et al. Sanderson. *Australian J. Chem* **69(12)**, (2016) 1402.

4. Ding R. L. et al. *Drug Deliv.* (2017), **24(1)**, 300.
5. Supported by the US Department of Defense [FA9550-14-1-0330 & W911NF-14-1-0139].

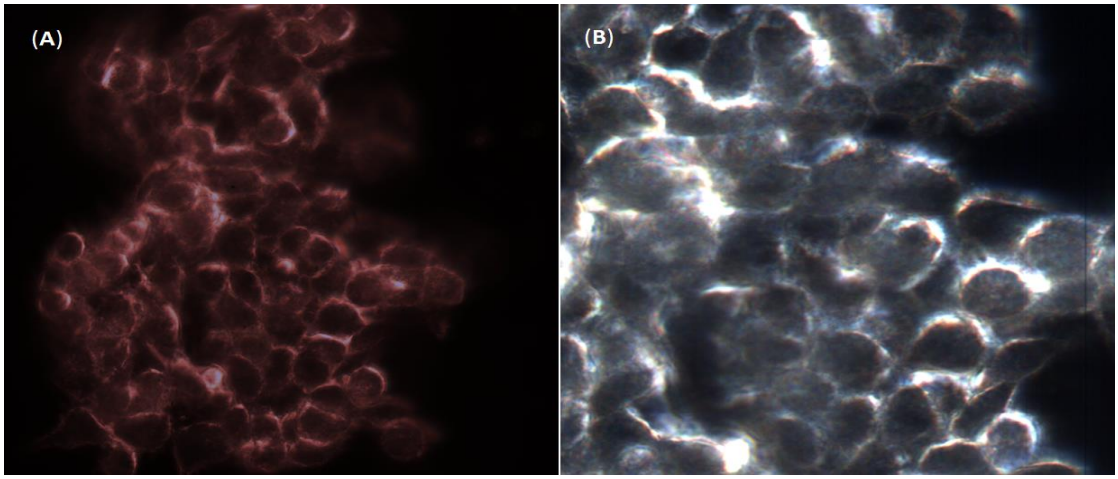

Figure 1: Optical image (A) and an image of Hyperspectral scan (B) of the same site on SW480 cell monolayer.

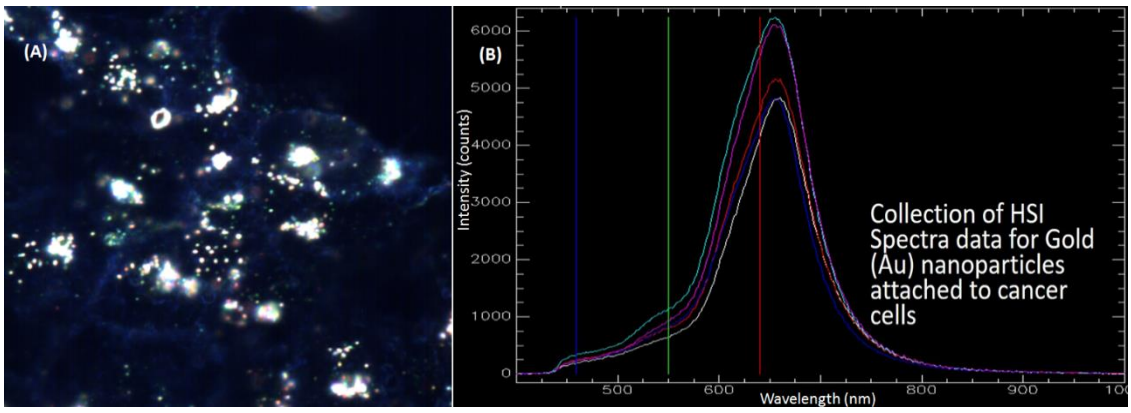

Figure 2: HS image of AuNPs and SW480 cells (A) and a collection of the Hyperspectral profiles of the nanoparticle on the same specimen (B). λ_{max} is constant while I_{max} drops due reduced pixel counts.

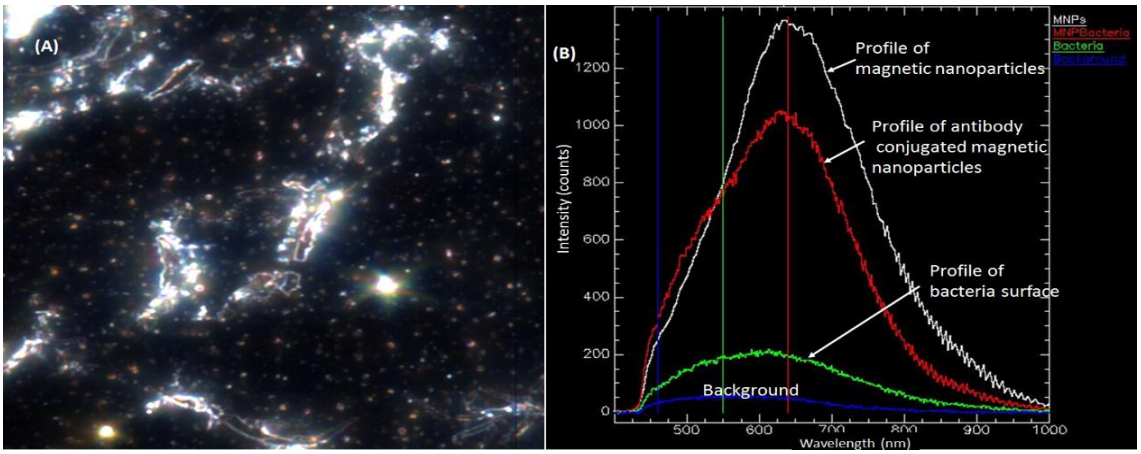

Figure 3: Hyperspectral image (A) and overlay of spectral profiles of antibody-conjugated magnetic particles and bacteria (B). The bare NPs have the highest peak intensity. This drops significantly once the NPs are functionalized. The bacteria produce the lowest spectrum intensity. This shows that by HSI, it is possible to indicate the functionalization of NPs and the interaction between particles and cells.

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