Micro-Raman spectroscopy study of Iron Deficiency Anemia and Beta-thalassemia

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Introduction

- Optical Tweezers – A powerful tool for the manipulation of microscopic particles without any mechanical contact.
- Tightly focused laser beam creates gradient optical force that traps the particles.
- Raman spectroscopy – highly reliable optical technique for investigating blood components such as red blood cells, platelets etc.
- Optical tweezers can be combined with Raman spectroscopy for biochemical evaluation of single, live cell.
- Iron Deficiency Anemia (IDA) and Beta-Thalassemia are two most common types of microcytic anemia. A micro-Raman spectroscopy study of the above cells are presented in this poster.
- Anemia is a condition marked by a decrease in the number of red blood cells, which is usually linked to a drop in hemoglobin levels or morphological alterations in red blood cells.

Objective

1. Raman Tweezers Technique to compare single live RBC spectra of Iron Deficiency Anemia and Beta-Thalassemia with healthy RBCs.

Experimental/Methodology

- The whole blood was centrifuged for 5 minutes at a rate of 3000 rpm for separating packed RBCs (PRBCs), platelets and blood plasma.
- The packed RBCs obtained from the healthy as well as patients were suspended in blood plasma for trapping a single cell.
- Laser power used for the experiment is 7mW.
- Raman spectra were recorded by focusing 785 nm laser beam on the RBC using a 100x oil immersion objective (NA 1.3) at an integration time of 60 s.
- Spectra were recorded using iHR-320 spectrograph having liquid nitrogen cooled CCD.

Results and Discussion

- The microcytic RBCs of Iron Deficiency Anemia has decreased intensity of pyrrole breathing mode region (752 cm⁻¹).
- The relative intensity of Pyrrole deformation mode region (heme) (674 cm⁻¹) was reduced in IDA.
- In the case of IDA the intensity of Tyrosine peaks are more intense compared to Thalassemia and normal.
- Normal RBC and the Beta thalassemia RBC has got almost similar intensity in pyrrole breathing mode region. So IDA can be discriminated by comparing the peak intensities of porphyrin breathing mode region and tyrosine bands.
- The oxygenation marker peaks (1222, 1561 and 1636 cm⁻¹) relative intensities are reduced in IDA and beta thalassemia.
- The relative intensity of Fe-O2 stretch (565 cm⁻¹) was reduced in IDA and beta thalassemia.

Conclusions

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References