







Multispectral imaging of healthy and diseased red blood cells using confocal microscopy

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ABSTRACT: Red blood cells (RBCs) alterations can cause very severe blood diseases. In thalassemia, the production of hemoglobin polypeptide chains is unbalanced. This causes no symptoms or small blood count alterations in patients with less important genetic mutations, or severe anemia and high morbi-mortality in the most severe cases. Traditional techniques do not perfectly discriminate among different thalassemia degrees and most patients need to undergo genetic studies. Authors have tried alternatives to analyze RBCs, collecting their emission and absorption spectral traits with spectrometers and spectrofluorometers. Multispectral imaging with confocal microscopy allows capturing both tridimensional and spectral information. In this study we are using confocal spectral imaging to analyze RBCs from patients with different degrees of alpha-thalassemia and iron deficiencies. The instrument, a Leica TCS SP8 confocal microscope has high sensitivity hybrid cameras, a diode laser and a white laser combined with an acoustic-optic tunable filter. RBCs exhibited autofluorescence when excited at 405 nm. The mean emission intensities at 502 nm, 628 nm and 649 nm allowed us to discriminate between different etiologies.

Keywords: multispectral imaging, confocal microscopy, thalassemia, red blood cell.









