

# **A New FLIRR Technique to Investigate the Redox Ratio in Live Cancer Cells and Tissues**

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Fluorescence lifetime imaging is a sensitive technique to investigate endogenous molecules NADH, FAD and tryptophan (Trp) in living cancer cells and tissues. The characterization of these endogenous molecules help us to understand the heterogeneous distribution of the metabolic signals or mapping of the metabolic signals in cancer specimens. The traditional intensity-based redox ratio includes intensity artefacts due to differential absorption and scattering in tissues and usage of various average excitation intensity levels at different depths. We developed a FLIM assay, fluorescence lifetime redox ratio (FLIRR), based on discrete ROIs (2x2 pixels), which mirror intensity-based morphology and measures the heterogeneous environment of the lifetime distribution in the prostate cancer cells. Glucose uptake and glycolysis proceeds about ten times faster in cancer than in non-cancerous cells or tissues. Therefore, we assessed the glycolytic activity in prostate cancer in comparison to normal cells upon glucose stimulation by analyzing the NADH and Trp lifetime distribution and efficiency of energy transfer (E%). Furthermore, we treated prostate cancer cells with Doxorubicin. Increase in NADH a2% as an indicator of increased glycolysis in Prostate cancer cells and increased E% between Trp and NADH was seen upon glucose stimulation for various time durations.