Spatio-temporal dynamics and Metabolic Alterations of P53 Upon DNA Damage

For biological reaction-diffusion systems, live single cell spatial-temporal analysis of protein dynamics provides a mean to observe stochastic biochemical signaling which may lead to better understanding of cancer cell invasion, stem cell differentiation and other fundamental biological processes. This talk will describe methodologies used to investigate p53 activity and alteration of the metabolic pathway upon DNA damage. p53 is a tumor suppressor protein that regulates target genes involved in DNA damage migration and repair. If cells become stressed due to DNA damage, p53 will form tetramers in specific chromatin sites and active genes that trigger cell cycle arrest and/or apoptosis. We used the Number and Molecular Brightness (N&B) method to map aggregation processes in the entire cell upon DNA damage with cisplatin (a chemotherapy agent); thus revealing the spatial distribution of events, the site of tetramer formation and the time sequence of the aggregation events with quantitative information about the distribution and size of any intermediate aggregates that are formed. To gain information regarding fast dynamic processes we use laser beam line scanning on a conventional confocal microscope to reveal the transient binding dynamics across the nucleus. Given that p53 has a dual role in promoting oxidative phosphorylation (oxsphos) and glycolysis upon cellular stress and may play an important role in normal growth, development, and tumor suppression, we investigated the effect of metabolic changes upon DNA damage under the same treatment and tested if the concentration of p53 influences the balance between apoptosis and DNA repair. We used the fluorescent lifetime imaging microscopy (FLIM) phasor approach to detect changes in oxsphos and glycolysis. The FLIM/Phasor data show that low concentrations of p53 is enough to trigger glycolytic response in the nucleus of the cells upon DNA damage with cisplatin and under high expression levels, a new lifetime phasor is detected. This new lifetime correlates with dead phenotype cells and may be a new "apoptotic" lifetime signal. Overall, our findings demonstrate that by multiplexing these techniques we have the ability to spatially and temporally quantify p53 activation and map p53’s influence in the metabolic pathway. This work is supported in part by NIH grants P50 GM076516 and P41 GM103540.

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Her current research interest focuses on the extracellular microenvironment regulating spatial-temporal biochemical dynamics and correlating it to cellular diversity in the area of cancer.