Cell cycle effects on Androgen Receptor Functions

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Short Abstract — Single-cell based studies on nuclear receptor (NR) mediated gene activation demonstrate a range of responses to environmental and physiological stimuli not previously appreciated by population-based studies. During the transcriptional activation process, AR changes spatial organization within the cell, including nuclear translocation and organization into subnuclear speckles. We identify and categorize subpopulations of AR-responsive cells during a time- and concentration-based series defining factors that are linked to responsive vs. non-responsive cells, and may indicate that cell cycle phase influences sensitivity to ligands.

***Keywords* — Cell cycle, Androgen Receptor, gene activation, single-cell level, microscopy.**

I. Purpose

Single-cell based studies on nuclear receptor (NR) mediated gene regulation demonstrate a range of responses to environmental and physiological stimuli not previously appreciated by population-based studies[1,2]. Androgen Receptor (AR) is critical for male sexual differentiation and development, bone mineral density, and muscle strength and mass in the adult. It is also related to diseases such as Androgen Insensitivity Syndrome and is important for prostate cancer development, survival, and growth [3,4]. Utilizing imaging modalities we can visualize single cells and measure distinct cellular responses to ligand in an androgen receptor system and correlate these responses to sources of heterogeneity.

During the transcriptional activation process, AR changes spatial organization, a microscopically visible process, including translocation from the cytoplasm into the nucleus and organization into subnuclear speckles [4]. To examine these ligand-based population shifts, the variation within the AR subcellular trafficking, and synthesis of target gene mRNA we developed a quantitative biological approach utilizing a stably-expressing GFP-AR HeLa and prostate cancer cell lines. We identify and categorize subpopulations of AR-responsive cells during a time- and concentration-based series defining extrinsic factors, specifically cell cycle, that are linked to responsive vs. non-responsive cells. We can then directly explore the correlation of gene activation to cell cycle phase by utilizing mRNA Fluorescent in situ Hybridization (mRNA FISH) and an established cell cycle phase identification technique [5]. Further analysis, both experimentally and mathematically, of this data will allow us to quantify individual cellular responses and the extrinsic and intrinsic variation between these responses from a seemingly homogenous population.

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