DUAL FLIM/PLIM IMAGING IDENTIFIES HYPOXIC REGIONS RESISTANT TO PI3K-PATHWAY TARGETED THERAPIES IN PANCREATIC CANCER

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ABSTRACT: By applying novel intravital imaging techniques to dynamically monitor pathway activity upon therapeutic inhibition, we are assessing PI3K-pathway targeted therapeutic resistance within the hypoxic microenvironment of pancreatic ductal adenocarcinoma (PDAC) [1]. Hypoxia is a negative prognostic factor in PDAC, known to increase radioresistance, chemo resistance, angiogenesis and metastasis. Here we demonstrate pronounced resistance in hypoxia to three clinically relevant inhibitors of the PI3K pathway; Rapamycin (mTORC1 inhibitor), NVP-BEZ235 (dual PI3K/mTORC1 inhibitor) and AZD2014 (mTORC1/2 inhibitor) [2]. Further to the clinical problem, we have mapped tumour hypoxia by both immunofluorescence and phosphorescence lifetime (PLIM) imaging techniques and here have shown that these hypoxic regions move sporadically around the tumour. Using advanced Akt Förster Resonance Energy Transfer (FRET) biosensors and state-of-the-art multiphoton imaging, we will demonstrate improved efficacy for dual PI3K/mTOR inhibitors against this hypoxia-induced resistance.

Within our well-established mouse models of PDAC, we have expressed an optimised Akt-FRET biosensor [3]. To date, in vitro modeling has involved three-dimensional invasion/proliferation assessment using organotypic assays and assessment of drug kinetics through live cell fluorescence lifetime (FLIM)-FRET imaging and standard molecular techniques. Progressing to in vivo intravital imaging, we are dynamically monitoring the therapeutic effect of targeting the PI3K–Akt–mTOR signaling axis within the hypoxic microenvironment, using novel dual FLIM/PLIM imaging techniques. Here we are able to assess novel combination therapies using hypoxia-activated prodrugs (i.e. TH-302) and angiogenics, with our dual PI3K pathway inhibitors, to improve treatment efficacy [4]. Further to our in vivo fidelity, we have developed a novel Akt-FRET biosensor mouse for real time analysis of drug kinetics in the native disease state.