Clinical Validation of a live-cell phenotypic biomarker: based diagnostic assay for the prediction of adverse prostate pathologies

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Introduction and Objective: Prostate cancer accounts for over 26% of total cancer cases in the United States. Current screening and diagnostic approaches lack the sensitivity to objectively assess the aggressiveness of tumors. To address this issue, a diagnostic assay was developed to differentiate indolent from aggressive tumors, objectively risk stratify patients and predict adverse pathology. Here we describe a diagnostic platform that is based on the measurement of a panel of phenotypic and molecular biomarkers in live biopsy-derived cells. Combining these tools, we are able to distinguish benign from malignant cells, local aggressiveness and invasiveness, respectively.

Methods: This clinical validation study was done on 54 prostate cancer samples (n=256) obtained at the time of radical prostatectomy. Patient cells were cryopreserved as ex vivo (up to 7 h) to enable live-cell, label-free imaging of multiple phenotypic biomarkers. Cells were incubated with an automated imaging device. Data were objectively quantified by machine vision algorithms to evaluate cellular behavior, and machine learning algorithms to standardize measurement.

Results: The developed predictive dynamic performance of the assay, measuring the Local Adverse Pathology Potential (LAPP) and Metastatic Adverse Pathology Potential (MAPP), report on the local aggressiveness and invasiveness, respectively. LAPP is able to distinguish benign from malignant cells, with greater than 90% sensitivity and specificity. LAPP and MAPP metrics can also predict the likelihood of six different adverse clinical pathologies with high accuracy as characterized by Receiver Operator Curves with Area Under the Curve (AUC) values >0.8. Conclusion: This novel live-cell phenotypic assay can quantitatively risk stratify patients with high accuracy as characterized by Receiver Operator Curves with Area Under the Curve (AUC) values >0.8.

Live cells are harvested from fresh samples. A biopsy surgical samples are collected and processed into single cell cultures. B) Extracellular Matrix (ECM) microfluidic devices are used to measure cell-cell interactions in the ECM environment for cell survival. C) Microfluidic devices, used in conjunction with ECM to promote cell survival, automate and standardize biomarker measurement. D) Growth curve of cell line from patient sample shows cells are available for analysis on day 2.

Novel diagnostic platform measures phenotypic (by optical and molecular) biomarkers on live cells harvested from patient tumor samples. A) Flow diagram illustrates automated process of sample procurement, sample processing, biomarker measurement, algorithmic processing and generation of predictive measurements. B) Biophysical and molecular biomarkers are automatically measured on subsequently fixed samples. Diagram of example biomarkers measured with single cell resolution.