UTILIZING ECTOPIC HSP90 EXPRESSION TO DIAGNOSE BREAST CANCER AT THE POINT-OF-CARE USING FLUORESCENCE MICROSCOPY

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Although pathological examination serves as the gold standard for breast cancer diagnosis, it requires labor-intensive sample preparation and time-consuming evaluation, resulting in long turn-around time and extensive infrastructure. We have developed a simple molecular imaging platform that can quickly assess patient's samples and provide a molecular signal to reflect disease pathology as an alternative to traditional pathology, particularly for applications in low resource settings. We identified Heat shock protein 90 (Hsp90) as a molecular target to diagnose breast cancer as it is overexpressed on the surface of all breast cancer cell subtypes to orchestrate stress response to cancer formation. Based on this feature, we have established a non-invasive and rapid molecular imaging approach to quantify Hsp90 expression on breast tissue biopsies using a FITC tethered Hsp90 inhibitor (HS-27) that binds to surface Hsp90 of breast cancer cells. A wide-field, high resolution, handheld fluorescent microscope referred to as the Pocket Mammoscope has been developed to perform rapid non-contact Hsp90 fluorescent imaging of entire tissue biopsies at point of care.

The Pocket Mammoscope uses a concentric excitation blue LED source (470±20 nm) a band-pass emission filter (534±26nm) for HS-27 fluorescence emission and a CMOS detector for imaging(Figure 1A). It is capable of 3-52X magnification and can perform both wide-field and high-resolution imaging by changing the working distance using a user-controlled slider mechanism on the body of the scope. In wide-field imaging mode, the Pocket Mammoscope achieves a FOV of 35 mm and an optical resolution of 24.8 μm while high-resolution mode achieves a FOV of 8.5 mm and an optical resolution of 8.77 μm. This dual imaging mode of Pocket Mammoscope allow us to image entire region of a core needle biopsy in one snapshot using the wide-field mode and zoom in to the regions of interest using a high-resolution mode (Figure 1B).

In our pilot clinical studies, we also demonstrated the feasibility of using the Pocket Mammoscope for Hsp90 fluorescent imaging on patient breast core needle biopsies. We showed that HER2+ biopsy has significantly higher HS-27 signal compared to benign breas ttissues (Figure 1C). To examine the sensitivity and specificity of Hsp90 imaging, we used features from the image in a Gaussian support vector machine (GSVM) classifier. A Receiver Operating Characteristics (ROC) curve shows specificity (100%) and sensitivity (86%) of Hsp90 for distinguishing malignant tumor biopsies (n = 27) from benign biopsies (n = 10) (Figure 1D). We are further optimizing our Hsp90 staining protocol and prototyping an automated staining platform to ensure uniform and consistent staining and washing of clinical biopsy samples as a way to improve sensitivity. By integrating an automated staining platform with Pocket Mammscope, our diagnostic platform could ultimately serve as an alternative to traditional pathology at a point-of-care setting.
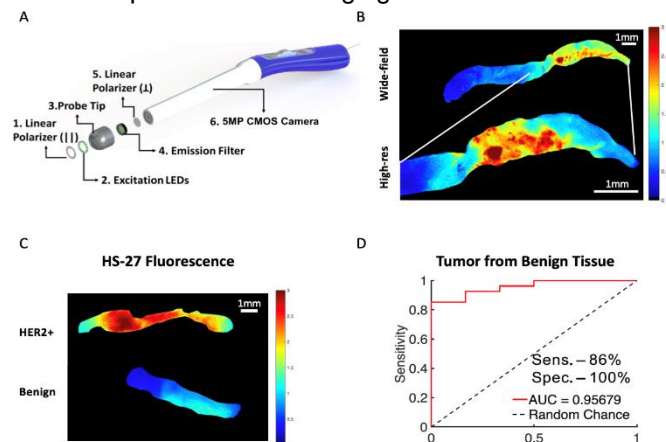

Figure 1 – (A) Pocket Mammoscope: (1) first linear film polarizer (2) concentric illumination with LEDs (470±20 nm) (3) aluminum holder (4) emission filter (535±26nm) (5) second linear film polarizer. (B) Representative image of cancerous breast biopsy captured by Pocket Mammoscope in wide-field (left) and high-resolution (right) modes. (C) Representative image of patient breast biopsies captured by Pocket Mammoscope: HER2+ (top) and Mammoplasty (bottom). (D) HS-27 ROC for distinguishing malignant tumor from benign tissue based on the top 1% of pixels