Background and Objectives

Stromal biology has been poorly studied compared to the cancer cell compartment and the immune cell population, until recent findings highlighted the indispensable influence of stromal reaction to the efficacy of treatment including immunotherapy. Collagen fibers are the major structural extracellular matrix component in breast and other tumours, and increased stromal collagen fibers have been found to facilitate breast tumour formation, invasion, and metastasis.

Materials & Methods

Stromal remodeling is featured by collagen realignment in the stromal compartment, collagen fibers and basal membrane, which can be identified, quantified and visualized by state-of-the-art stain-free pathology imaging technologies Second Harmonic Generation and Two Photon Emission (SHG-TPE). We profiled 388 triple-negative breast cancer (TNBC) patients with 68 SHG-TPE parameters and correlate with clinicopathological parameters.

Results

Among all the parameters, high Collagen Fiber Density (CFD) showed marginal association with better disease free survival and overall survival (DFS p=0.047; OS p=0.044). Furthermore, we developed algorithms to further differentiate the collagen fibers into “aggregated collagen fibres” and “thin collagen fibres” based on the complexity and texture of the fibers. Interestingly only aggregated CFD is associated with better prognosis (DFS and OS, p=0.014; p=0.033) but not thin CFD. On the other hand, if we consider the incremental 1 percent of aggregated/thin collagen ratio was associated with better DFS (HR 0.97, 95% CI 0.96-1.00, p=0.015). The addition of aggregated/thin collagen ratio to clinicopathological features significantly increased the prognostic value for DFS ($\Delta$LR$^2$= 8.4, p=0.004), compared to clinicopathological features alone (histological grade, tumour size and lymph node status).

Summary and Discussion

Our automated image analysis pipeline with SHG offers a novel platform to quantify stromal collagen of the tumor microenvironment to better predict the clinical outcome of patients with triple negative breast cancer. With breast cancer as proof-of-concept for our findings, we plan to apply and validate in other cancers, potentially translating to clinical applications in the imminent future. SHG is widely used to assist diagnosis of fibrosis disease. Our data showed potential application on cancer setting however validation with bigger cohort and other cancers are warranted. Further study might need to focus on prediction of treatment response such as immunotherapy.

Acknowledgement

This study was approved by the Singapore Centralized Institutional Review Board (IRB Ref: 2015/064F and 2017/0210), and supported by the Singapore National Medical Research Council Transition Award (NMRC/TA/401/2015), the SingHealth Duke-NUS Pathology Academic Clinical Program Building Clinician-Scientist grant (ACP PATH BCS 14 021), and the A*STAR Biomedical Research Council, National Medical Research Council Stratified/National Programme Office (SMP0201.200). The authors declared no conflict of interest.

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