



Biomarker Panel for Triple Negative Breast Cancer

Novel panels of epigenetic biomarkers for:

- Diagnosis of triple negative breast cancer (TNBC)
 - Sensitivity: 0.72; Specificity: 0.94; AUC 0.9
 - Accurate patient stratification to ensure TNBC patients are correctly treated with chemotherapy
 - Minimal residual disease detection in blood
- Predicting prognosis of patients with TNBC

Triple-negative breast cancers (TNBC) comprise a heterogeneous group of cancers, named as such because they do not express estrogen receptor (ER), progesterone receptor (PR) or herceptin (HER-2) and have varying prognoses. TNBC represents ~15-20% of all newly diagnosed breast cancer cases and is generally associated with poorer prognosis than non-TNBC. Whilst targeted therapies are available for other subtypes of breast cancer, such as herceptin for HER-2+ breast cancers, these are ineffective for TNBC for which chemotherapy remains the front-line treatment. Currently, immunohistochemistry is used to categorize breast cancer and, therefore, guide treatment decisions. However, this can be subjective based on staining levels for ER, PR and HER-2, generating potential high false-positive rates for non-TNBC subtypes, which results in ineffective treatment of these incorrectly defined tumours. There is a clear unmet need for a robust diagnostic method for identifying TNBC to supplement immunohistochemistry to improve treatment outcomes as well as for sensitive molecular blood biomarkers to monitor tumour relapse before the cancer spreads.

Furthermore, TNBC patients can be broadly divided into two distinct groups: those with a poor prognosis (3-5 years survival) and those with a good prognosis (survival >8-10 years). There is a need to be able to stratify patients into high and low risk in order to make informed decisions relating to disease management.

Garvan researchers performed genome-wide next generation DNA methylation profiling of formalin-fixed paraffin embedded (FFPE) triple-negative clinical DNA samples (using MBDCap-Seq).

A diagnostic panel of 282 TNBC-specific probes is capable of identifying TNBC

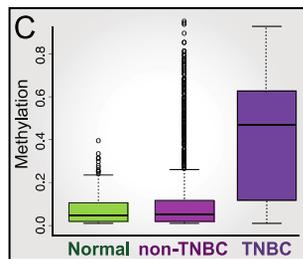
Specifications:

- Panel of 282 TNBC-specific probes
- Sensitivity: 0.72
- Specificity: 0.94
- AUC: 0.9

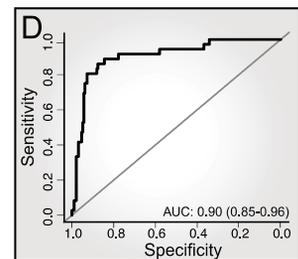
Training Set: TNBC n=37; non-TNBC n=193

Test Set: TNBC n=36; non-TNBC n=193

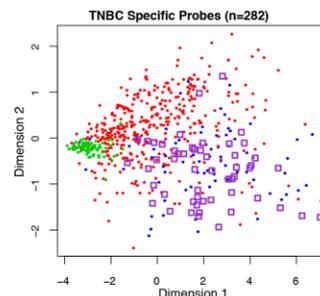
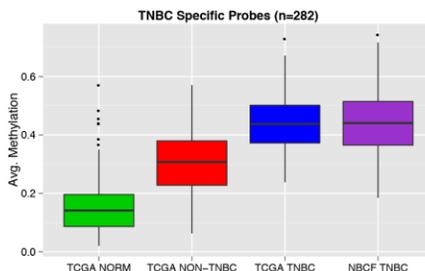
TNBC Specific Methylation



ROC Analysis



Correctly classifies 93% (49/53) samples from National Breast Cancer Foundation as TNBC



Competitive Landscape

Immunohistochemistry (IHC) (current primary method used to ER and PR status)

- Qualitative and is unreliable due to a lack of clarity regarding 'negative staining'.
- Arbitrary thresholds used for discrimination between positive and negative ER and PR status, ranging from 1-20% which vary between laboratories and rely heavily on interpretation by clinicians and pathologists.
- Reproducibility is an issue, with variation caused by tissue preparation, antibody choice and detection.
- A high false positive rate for non-TNBC subtypes, leading to incorrect treatment and poorer outcomes (e.g. the false positive rate for HER2 status has been predicted to be between 3-22%).
- FDA-approved IHC-based tests for HER2 overexpression are HercepTest (Dako) and Pathway (Ventana).

FISH-based methods (HER2 status only. Has surpassed IHC for routine testing):

- Qualitative and rely on interpretation, thereby resulting in inter-laboratory variability.
- They are complex and labour-intensive, with significantly higher failure rate, reagent cost, testing and interpretation time than IHC. Consequently, FISH is only used if the IHC staining is intermediate.
- FDA-approved tests include PathVysion (Abbott Molecular), INFORM (Ventana) and HER2 FISH parmDx (Dako).

Quantitative PCR methods (HER2 status only):

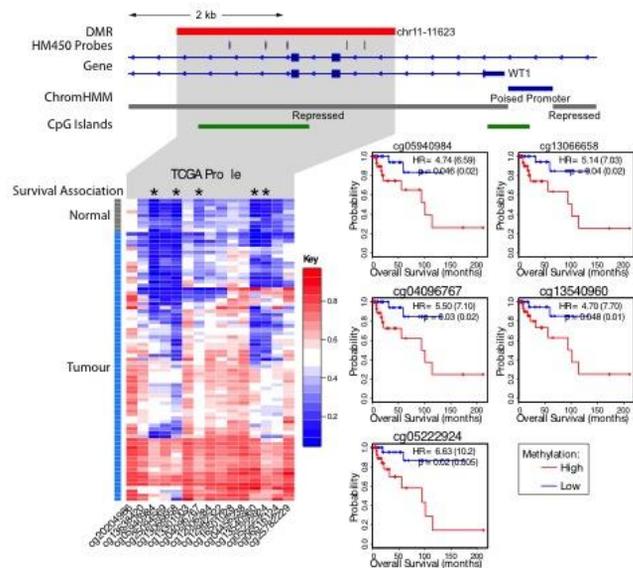
- Quantitative and less expensive than IHC and FISH
- Reliability is in dispute - some studies indicate IHC to be superior. Not currently adopted in clinical practice.

A prognostic panel of 190 probes shows excellent survival separation (Kaplan Meier plots)

High Methylation corresponds to poor survival.

Validated on an independent cohort of 53 TNBC (25 deaths; 28 alive).

- 4 probes predict high risk
- 3 probes predict low risk



Intellectual Property:

PCT/AU2015/050549

Publications:

Nature Communications, 2015

Opportunity:

Licensing or Collaborative Research

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