





# Tumour Infiltrating Lymphocytes (TILs) in triple-negative breast cancer: High Immunoscore is associated with pathological CR in patients

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### Background

- Neoadjuvant chemotherapy is widely used to downstage breast cancers prior to surgery.
- ▶ Pathologic complete response (pCR) rate is a strong predictor of outcome for breast cancer.

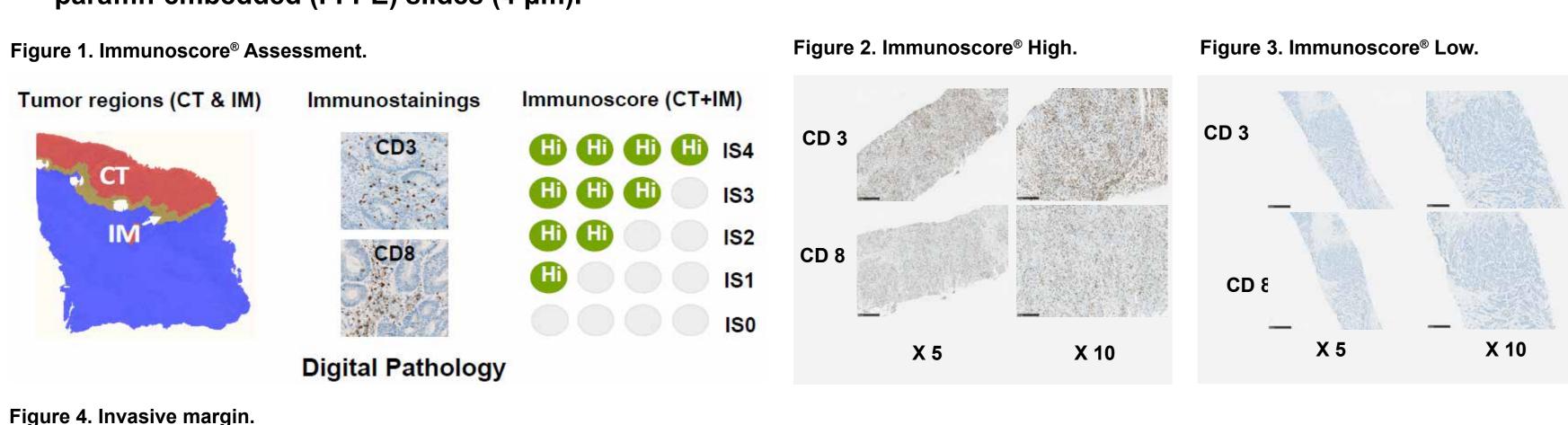
- ▶ The Immunoscore® assay is the first standardized immune-based assay for classification of cancer [Hermitte et al., 2016]. It assesses the host immune response by measuring intra- and peri-tumoral T cell infiltration in formalin-fixed paraffin-embedded (FFPE) tissue sections.
- Originally developed for colon cancer indication, it is intended to be widely used in solid cancer indications for diagnostic and prognostic purposes, as well as a pharmacodynamic biomarker during drug development processes. As a first clinical validation in breast cancer, we assessed the Immunoscore in a cohort of 103 breast cancer patients, that previously received neo-adjuvant chemotherapy.

### Methods

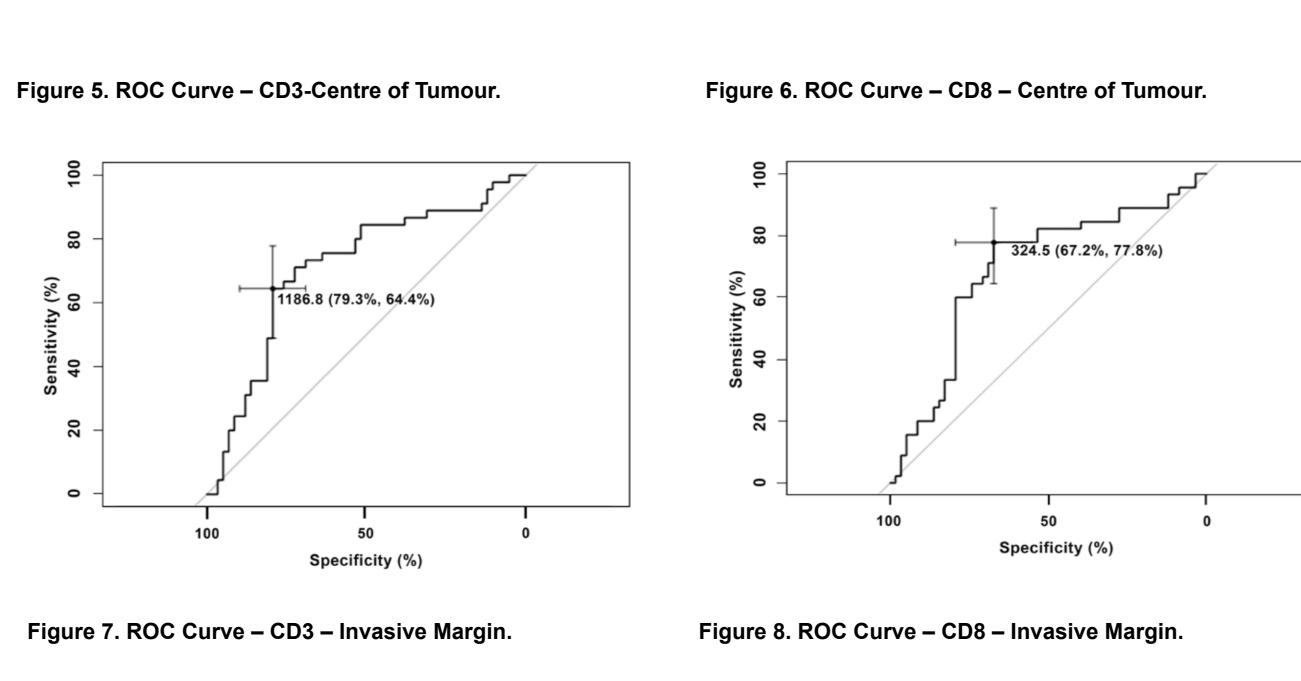
### Pathological and clinical assessment

- Clinical assessment of the primary tumour and lymph nodes was made using bi-dimensional caliper measurements of the primary tumour and axillary nodes.
- Sonographical assessments of the primary tumour and lymph nodes were performed regularly.
- Immunohistochemical staining was performed for ER, PR, HER-2 and Ki67.
- ▶ Fluorescence in situ hybridization (FISH) was used to confirm HER-2 positivity.
- We analyzed data retrospectively/prospectively on 103 breast cancer patients undergoing neoadjuvant chemotherapy.
- Pathological complete response (pCR) was defined as the complete disappearance of the invasive cancer in the breast and absence of tumour in the axillary lymph nodes.
- Ethics approval was obtained from Pharma-Ethics, Pretoria, South Africa (ethics committee working according to the South African Ethics regulations).
- NCSS software version 11 for Windows (USA) was used for statistical analyses.
- Outcome assessments: Associations of clinical and pathological characteristics including Ki67, CD8+ cytotoxic T cells and CD3+ T cells with pCR.
- All patients were treated with anthracycline and/or taxane-based neoadjuvant chemotherapy.

- In this retrospective analysis, 103 pre-treatment tumour tissue samples were analyzed by immunohistochemistry for density (cells/mm³) of T-cell subsets (CD3+,CD8+).
- ▶ CD3 and CD8 staining was performed using Benchmark® XT station on 2 consecutive formalin-fixed paraffin-embedded (FFPE) slides (4 µm).

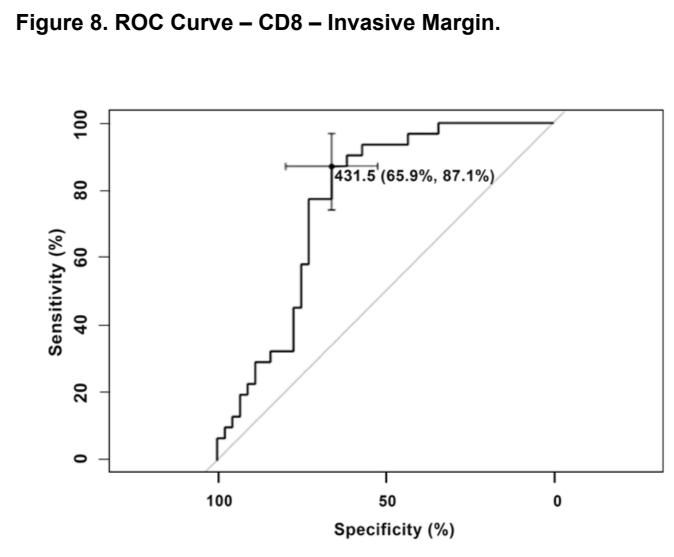


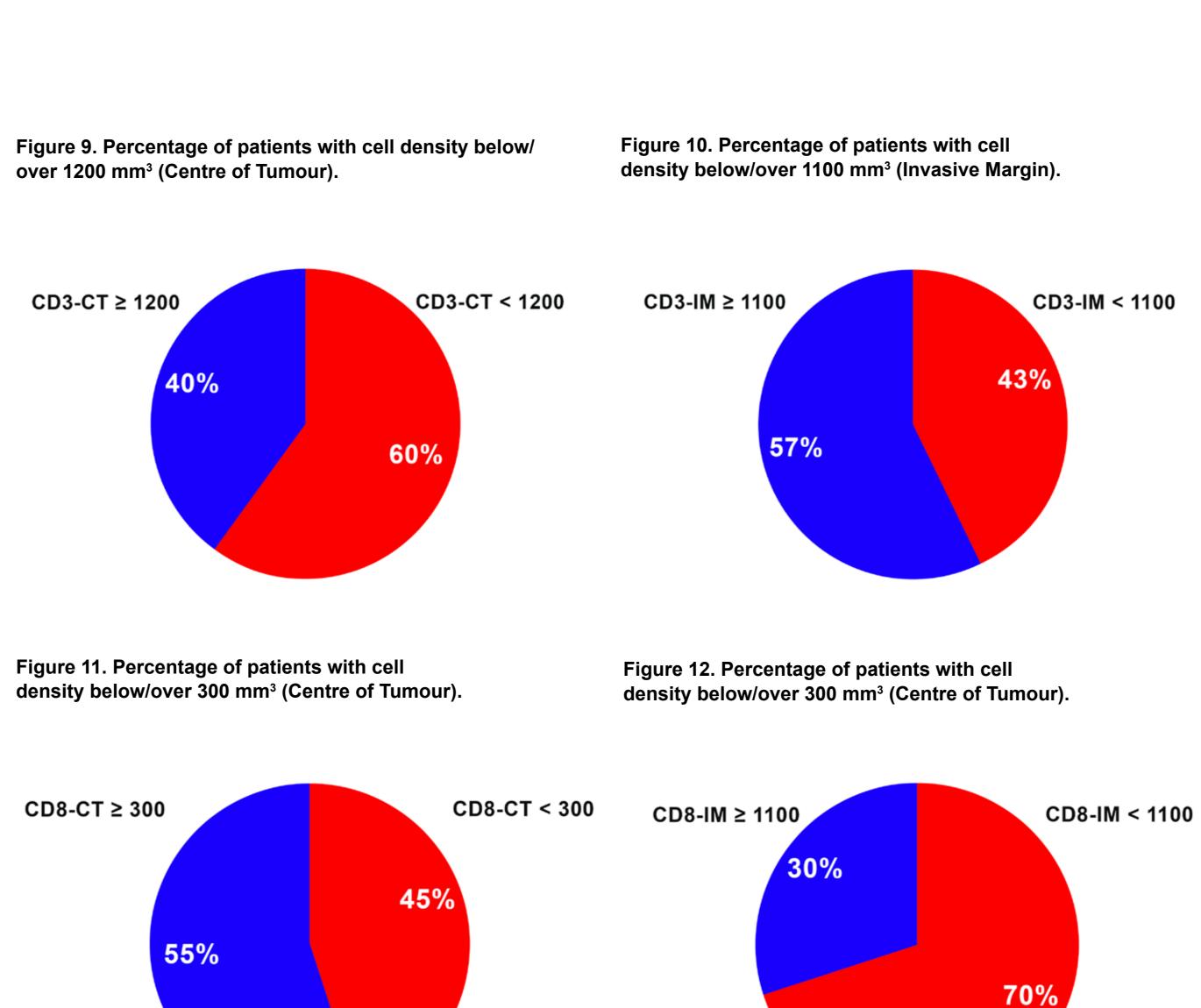
## Results The primary hypothesis was that higher levels of cell density between TNBC and Non-TNBC patients. analysis was used to determine the optimal cut-poin for Ki67, CD8+ cytotoxic T cells, CD3+ T cells and Fisher's exact or Chi-squared tests were used for the Logistic regression multivariate models included only variables that exhibited a univariate association

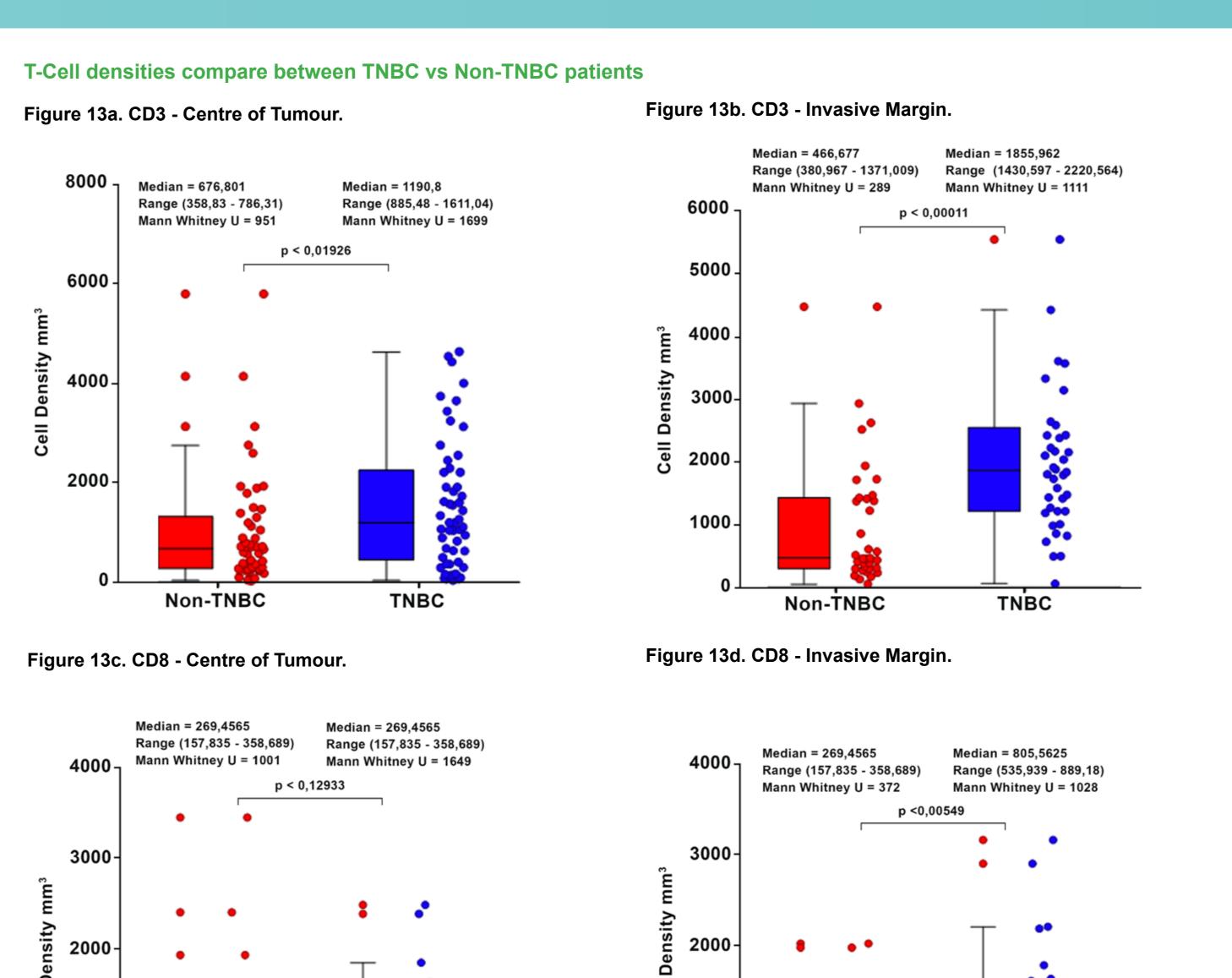


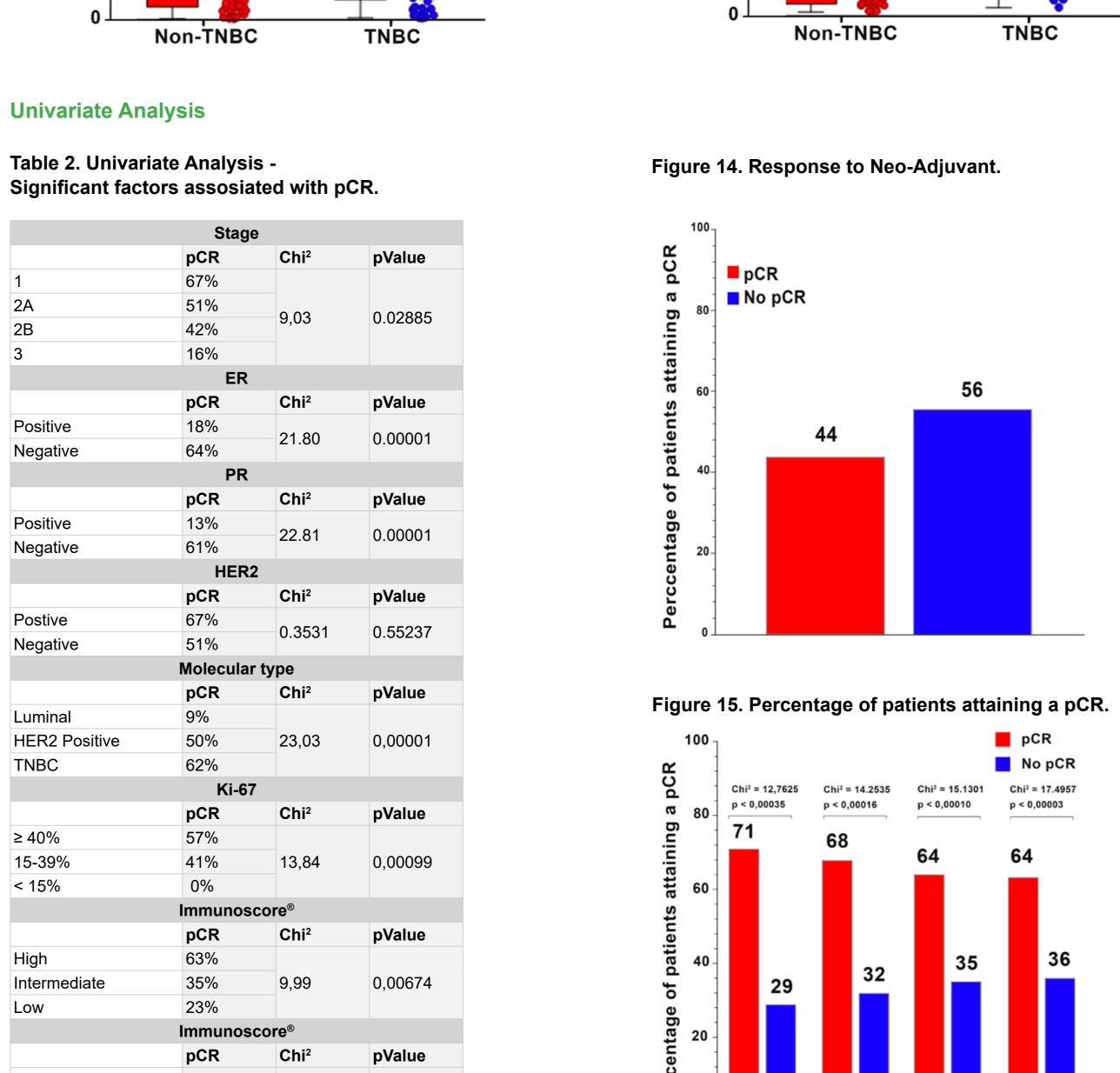
1093.7 (63.6%, 93.5%)

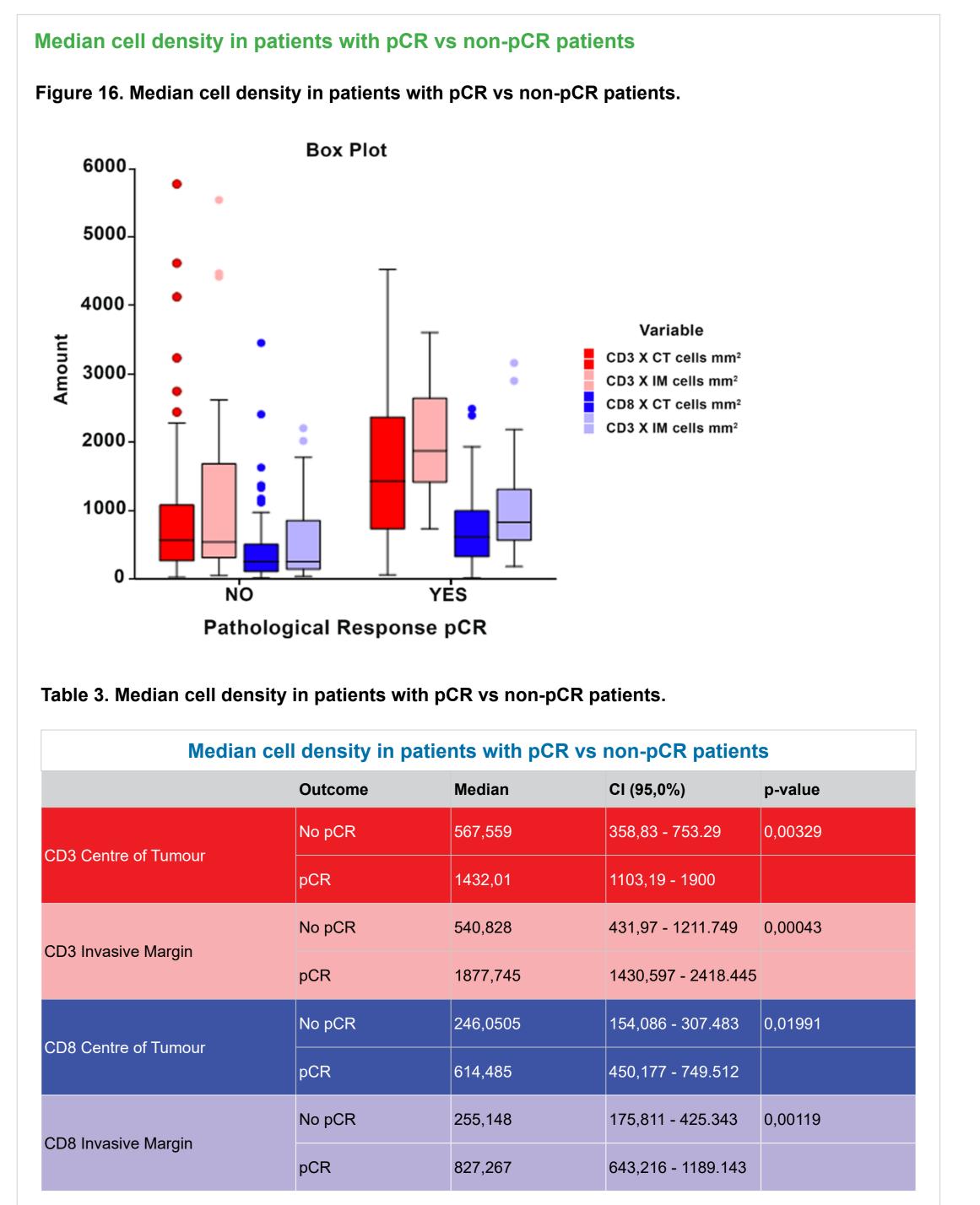
Specificity (%)

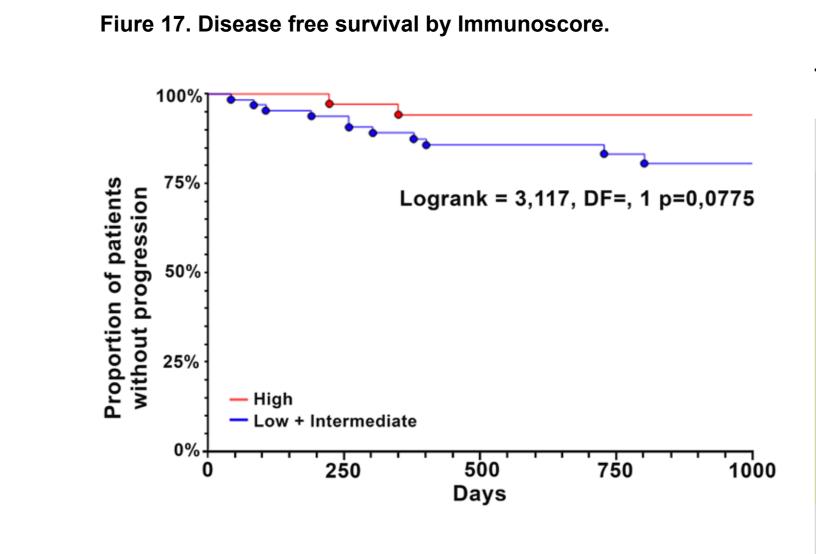


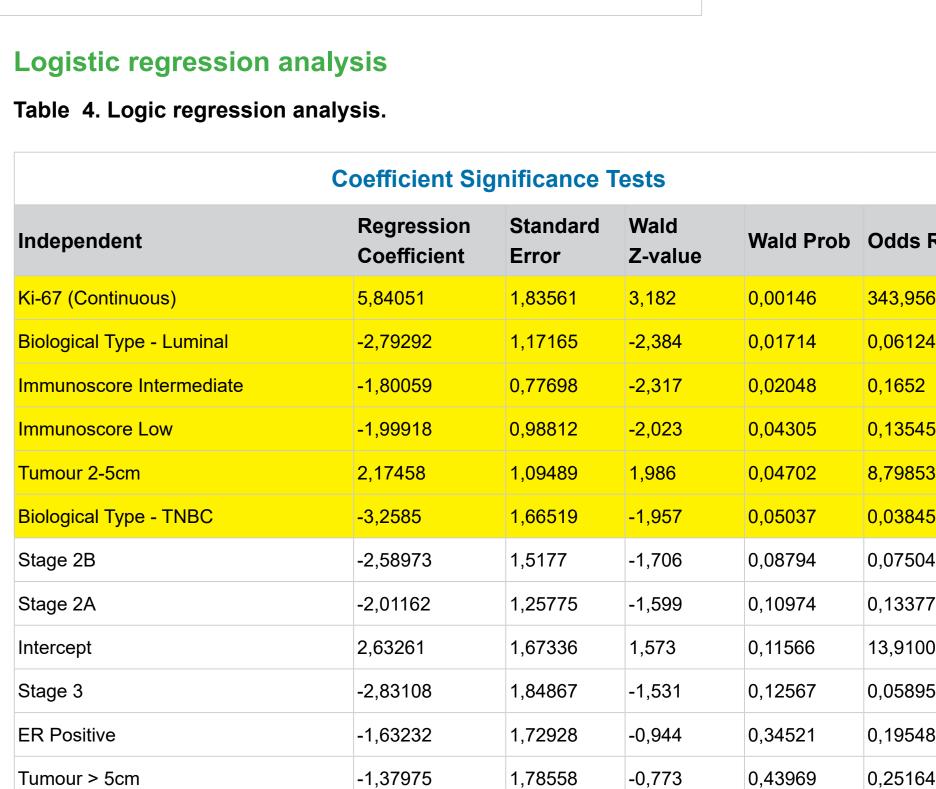








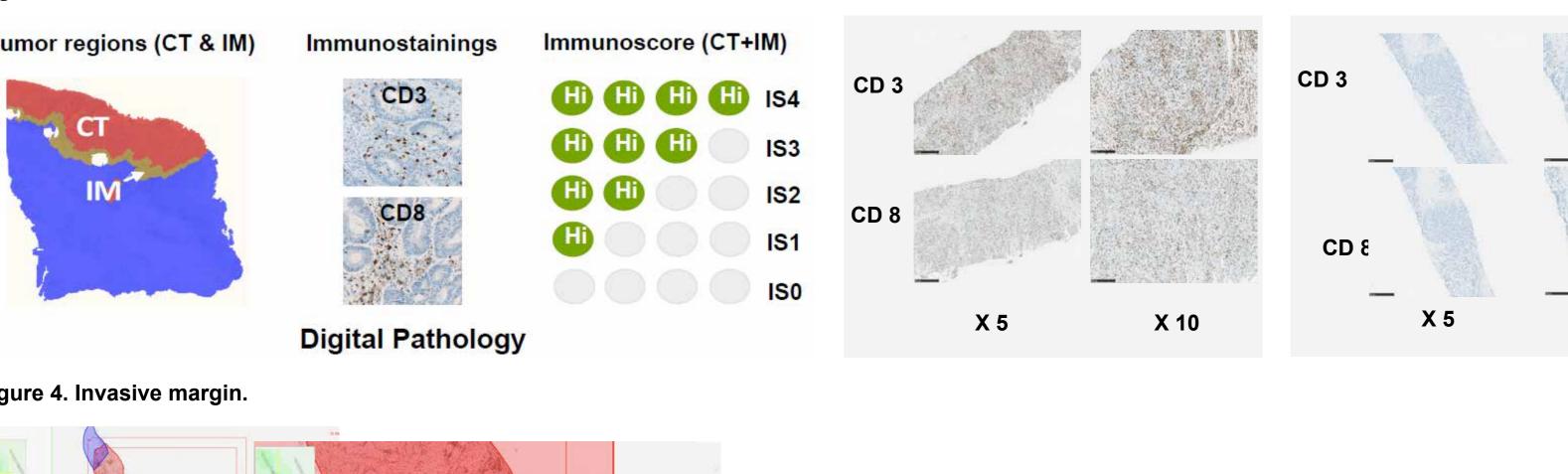


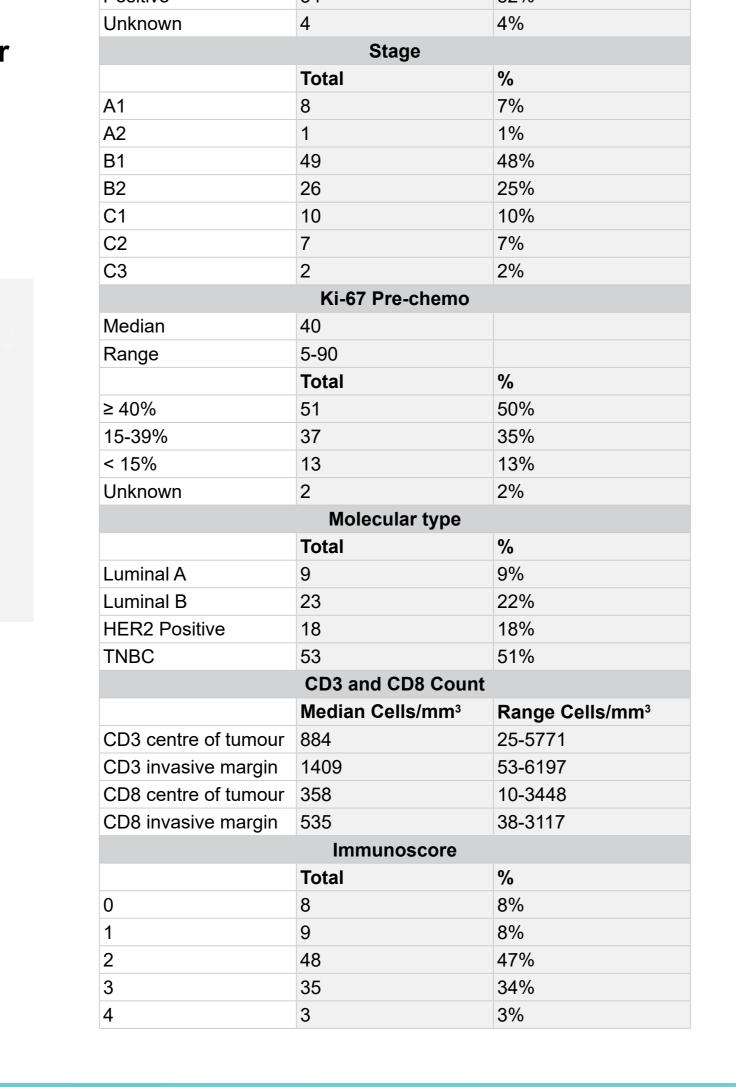




▶ Ki-67, Biological type, Immunoscore® and tumour size are independent prognostic factors of pCR in patients with early breast cancer undergoing neoadjuvant chemotherapy.

PR Positive





CD8+ cytotoxic T cells, CD3+ T cells and

Immunoscore®.

analysis of categorical variables.

used for statistical analyses.

**Table 1. Patient Characteristics.** 

with the dependent variable, pCR (p < 0.1).

NCSS software version 11 for Windows (USA) was

Patient Characteristics

Age (n=103)

overall prognosis, independent of anti-cancer

