
Supplementary information

**Spatial predictors of immunotherapy
response in triple-negative breast cancer**

In the format provided by the
authors and unedited

Annotation for supplementary material associated with *Spatial predictors of immunotherapy response in triple negative breast cancer*

Xiao Qian Wang¹, Esther Danenberg¹, Chiun-Sheng Huang², Daniel Egle³, Maurizio Callari⁴, Begoña Bermejo^{5,6,7}, Matteo Dugo⁸, Claudio Zamagni⁹, Marc Thill¹⁰, Anton Anton¹¹, Stefania Zambelli⁸, Stefania Russo¹², Eva Maria Ciruelos¹³, Richard Greil^{14,15,16}, Balázs Gyórfy^{17,18}, Vladimir Semiglazov¹⁹, Marco Colleoni²⁰, Catherine M. Kelly²¹, Gabriella Mariani²², Lucia Del Mastro^{23,24}, Olivia Biasi²⁰, Robert S. Seitz²⁵, Pinuccia Valagussa⁴, Giuseppe Viale^{20,26}, Luca Gianni^{4,28}, Giampaolo Bianchini^{4,8,28*}, H. Raza Ali^{1,27,28*}

¹ CRUK Cambridge Institute, University of Cambridge, Cambridge, UK

² National Taiwan University Hospital, College of Medicine, National Taiwan University and Taiwan Breast Cancer Consortium, Taipei, Taiwan

³ Department of Gynecology, Brust Gesundheit Zentrum Tirol, Medical University Innsbruck, Innsbruck, Austria

⁴ Fondazione Michelangelo, Milan, Italy

⁵ Medical Oncology, Hospital Clínico Universitario de Valencia, Biomedical Research Institute INCLIVA, Valencia

⁶ Medicine Department, Universidad de Valencia.

⁷ Oncology Biomedical Research National Network (CIBERONC-ISCIII), Madrid.

⁸ San Raffaele Hospital, Milano, Italy

⁹ IRCCS Azienda Ospedaliero-universitaria di Bologna, Italy

¹⁰ Department of Gynecology and Gynecological Oncology, Agaplesion Markus Krankenhaus, Frankfurt am Main, Germany

¹¹ Hospital Universitario Miguel Servet, Zaragoza, Spain

¹² Department of Oncology, Azienda Sanitaria Universitaria Friuli Centrale, Udine, Italy

¹³ Hospital Universitario 12 de Octubre, Madrid, Spain

¹⁴ 3rd Medical Department, Paracelsus Medical University Salzburg, Salzburg, Austria

¹⁵ Salzburg Cancer Research Institute-CCCIT, Salzburg, Austria

¹⁶ Cancer Cluster Salzburg, Salzburg, Austria

¹⁷ Semmelweis University Dept. of Bioinformatics, Budapest, Hungary

¹⁸ Cancer Biomarker Research Group, Research Centre for Natural Sciences, Institute of Enzymology, Budapest, Hungary

¹⁹ NN Petrov Research Institute of Oncology, St. Petersburg, Russia

²⁰ IEO, Istituto Europeo di Oncologia, IRCCS, Milan, Italy

²¹ Mater Private Hospital, Dublin and Cancer Trials Ireland Breast Group

²² Fondazione IRCCS - Istituto Nazionale Tumori, Milan, Italy

²³ IRCCS Ospedale Policlinico San Martino, UO Clinica di Oncologia Medica, Genoa, Italy

²⁴ Università di Genova, Dipartimento di Medicina Interna e Specialità Mediche (Di.M.I.), Genoa, Italy

²⁵ Oncocyte Corporation, Irvine, California, USA

²⁶ University of Milan, Milan, Italy

²⁷ Department of Histopathology, Addenbrookes Hospital, Cambridge, UK

²⁸ These authors jointly supervised this work

*Correspondence: bianchini.giampaolo@hsr.it ; raza.ali@cruk.cam.ac.uk

Table of Contents:**Supplementary Figures:**

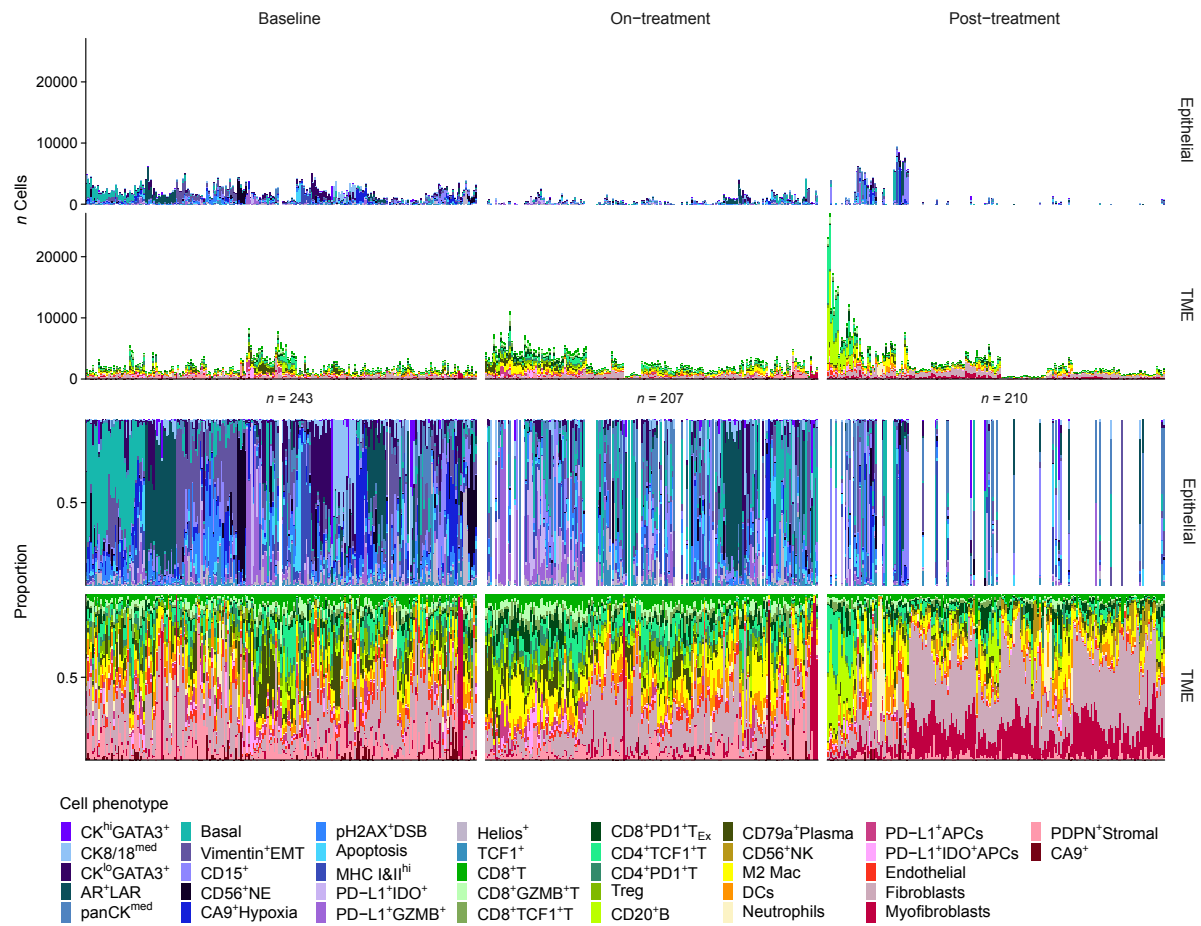
- **Supplementary Fig. 1** | Phenotypic composition of TNBC during neoadjuvant therapy (pg2)
- **Supplementary Fig. 2** | Cell counts per biopsy grouped by proliferative fraction quantiles (pg3)

Supplementary Tables:

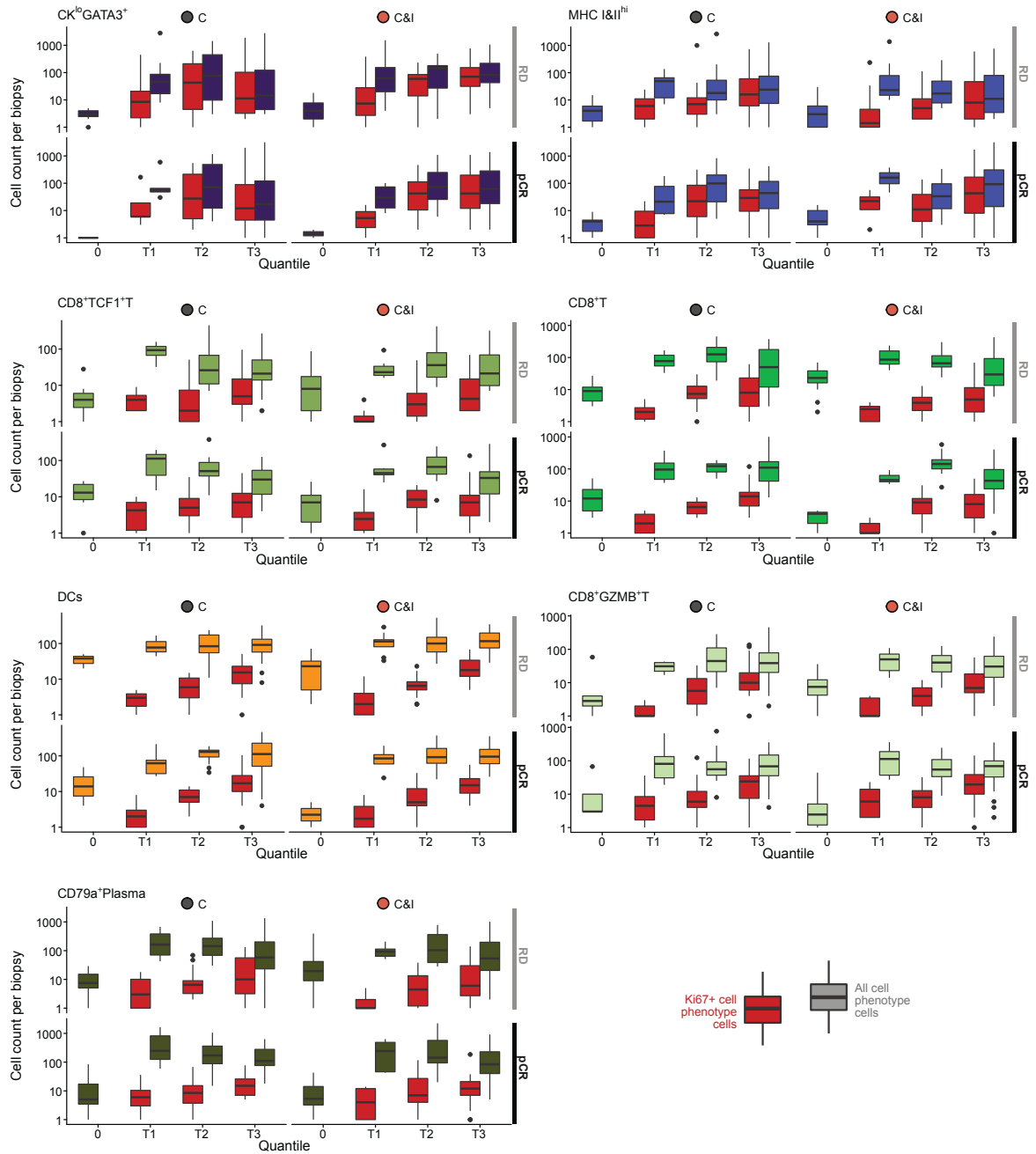
Table	Description
Table 1	Patient demographics and clinical characteristics - Please note that this is data for all 280 patients enrolled in the trial, of which 279 were analysed in this study.
Table 2	Patient exclusions, including reasons
Table 3	Paired counts - no. of samples available at multiple timepoints
Table 4	Counts used in most analyses - images containing invasive epithelial cells
Table 5	Counts by image type - no. of images containing only TME cells and number of images containing non-invasive epithelial cells
Table 6	List of ethics committees that approved of the NeoTRIPaPDL1 study
Table 7	Summary of cell phenotype densities across timepoints, treatment arms and response
Table 8	Point estimates, confidence intervals, false discovery rates and p-values of associations between cell densities and response
Table 9	Summary of cell-cell interaction values across timepoints, treatment arms and response
Table 10	Point estimates, confidence intervals, false discovery rates and p-values of associations between cell-cell interactions and response
Table 11	Point estimates, confidence intervals, false discovery rates and p-values of associations between proliferation fraction and response
Table 12	List of antibodies and metal conjugates in the panel

Abbreviations	Full
TME	Tumour microenvironment
B	Baseline
OT	On-treatment
PT	Post-treatment
C	Chemotherapy
C&I	Chemotherapy and Immunotherapy
RD	residual disease
pCR	pathological complete response
fdr	false discovery rate
estimate	point estimate derived from logistic regression
std.error	standard error
conf.low	lower bound of 95% confidence interval
conf.high	upper bound of 95% confidence interval

Each supplementary table is on its own sheet in the excel spreadsheet.



Supplementary Fig. 1| Phenotypic composition of TNBC during neoadjuvant therapy. Number and proportion of cell phenotypes by compartment (epithelial or TME) are depicted for each patient at each timepoint.



Supplementary Fig. 2 | Cell counts per biopsy grouped by proliferative fraction quantiles. Boxplots depict comparisons between absolute number of Ki67+ cells of specific phenotypes (red) next to number of total cells of specific phenotypes per biopsy per quantile shown in Fig. 3D. Boxes show 25th, 50th, and 75th centiles; whiskers indicate 75th centile plus 1.5x inter-quartile range and 25th centile less 1.5x inter-quartile range; points beyond whiskers are outliers. Numbers of patients for each group are the same as represented in Fig. 3D.