Supplemental methods:

Measurement of PD-L1 protein using QIF: PD-L1 protein was measured using the AQUA® method in FFPE TMA samples with the monoclonal antibody clone 5h1 (Dr. Lieping Chen, Yale University) as recently reported (Velcheti et al., 2014 [35]). Briefly, fresh TMA cuts were deparaffinized and subjected to antigen retrieval using tris-EDTA buffer (Sigma-Aldrich, St Louis, MO) with 0.05% Tween pH=9.0 and boiling for 20 min at 102 °C in a pressure-boiling container (PT module, Lab Vision). Slides were then incubated with 1% BlockAce (cat # BUF029, AbDSerotec, Oxford, UK) for 10 min at room temperature and incubated overnight at 4 °C with a solution containing primary PD-L1 antibody and a rabbit monoclonal anti-human pancytokeratin antibody (clone AE1/AE3, M3515; Dako Corporation, Carpinteria, CA). Sections were then incubated for 1 h at room temperature with Alexa 546-conjugated goat anti-rabbit secondary antibody (A11003; Molecular Probes, Eugene, OR, USA) in mouse EnVision amplification reagent (K4003, Dako). Cyanine 5 (Cy5) directly conjugated to tyramide (FP1117; Perkin-Elmer) was used for antibody detection. Prolong mounting medium (ProLong Gold, P36931; Molecular Probes) with 4′,6-diamidino-2-phenylindole (DAPI) was used to stain nuclei.

Quantification of TILs using QIF: Details of this protocol are presented in Brown et al. (Brown JR, et al. Quantitative Assessment of Tumor Infiltrating Lymphocytes is Predictive of Response to Neoadjuvant Chemotherapy in Breast Cancer, 2014, American Society for Clinical Oncology, Chicago IL, May 2014). Briefly, signal from different TILs subtypes expressing CD3, CD8 or CD20 were simultaneously measured

using QIF in different tumor compartments of FFPE TMA samples with a sequential multiplexed immunofluorescence protocol to detect epithelial tumor cells (cytokeratin, clone M3515, DAKO), T lymphocytes (CD3, clone E272, Novus biologicals, CO), cytotoxic T cells (CD8, clone C8/144B, DAKO), B-lymphocytes (CD20, clone L26, DAKO). Nuclei were highlighted using (DAPI). Secondary antibodies and fluorescent reagents used were goat anti-rabbit Alexa546 (Invitrogen), anti-rabbit Envision (K4009, DAKO) with biotynilated tyramide/streptavidine-Alexa750 conjugate (Perkin-Elmer); anti-mouse IgG1 antibody (eBioscience, CA) with fluorescein-tyramide (Perkin-Elmer), anti-IgG2a antibody (Abcam, MA) with Cy5-tyramide (Perkin-Elmer). Samples from human tonsil were used as positive controls in each run.

Validation of the prognostic effect of PD-L1 mRNA in available breast cancer datasets: To address the prognostic value of PD-L1 mRNA in publicly available breast cancer datasets we used the recently described tool SurvExpress (Aguirre-Gamboa R et al., 2013 [40]). Expression of PD-L1 mRNA was searched in the records from 30 available breast cancer mRNA expression datasets using as searching criteria CD274, PD-L1, B7-h1, B7h, PDCD1L1, PDCD1LG1 and PDL1. Results were obtained using the average score from available probe sets and using the original quantile-normalized format. Statistical analyses and graphical output were set using available dataset endpoints to obtain 2 maximized risk groups. Accessions to the server were between December 2013 and January 2014.