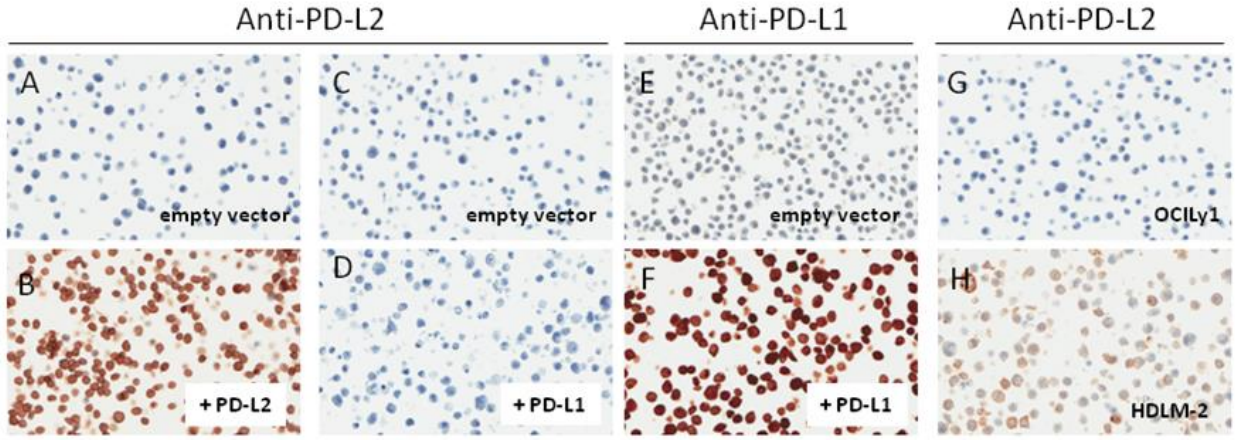
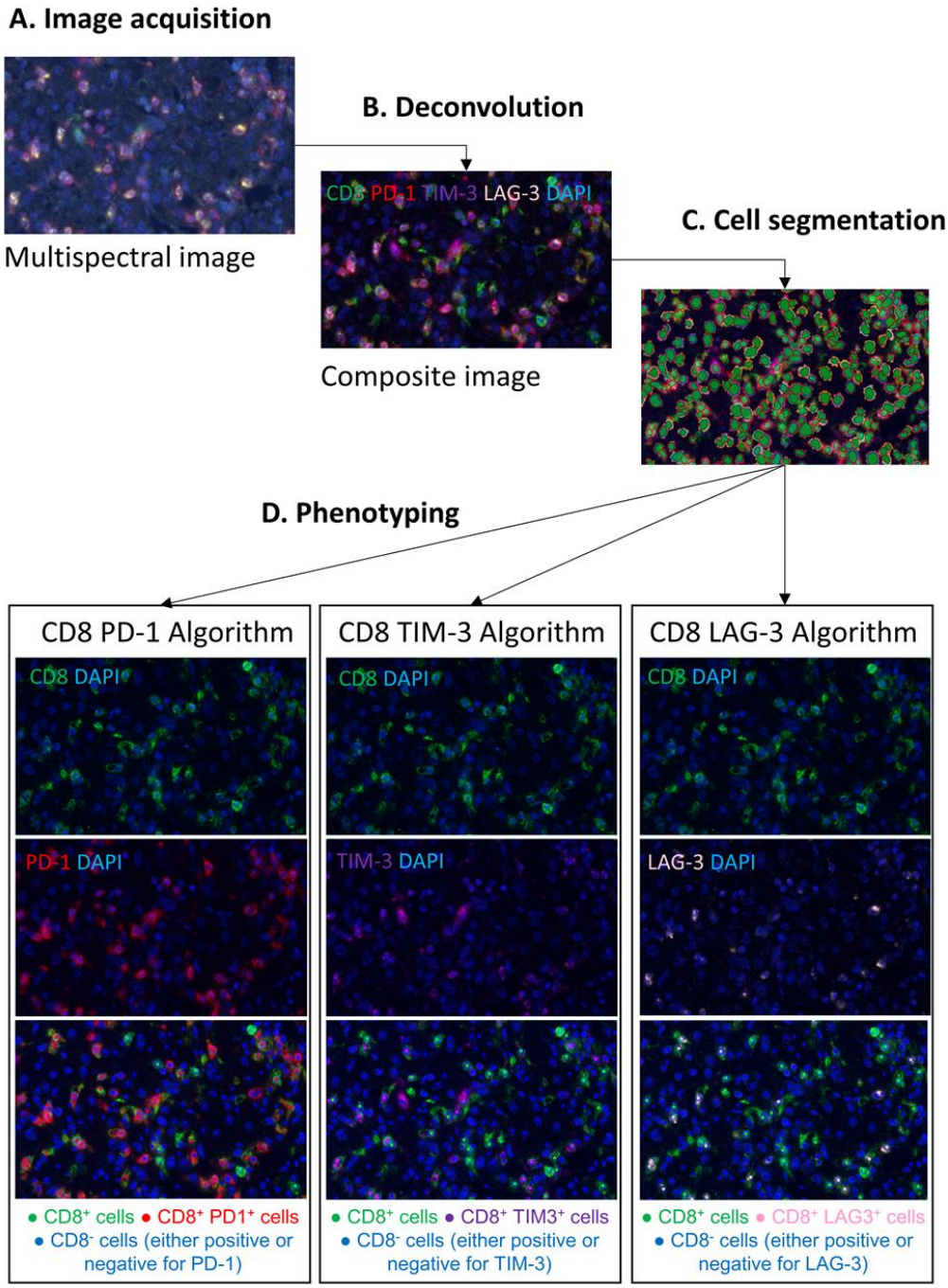
Supplementary Figures

Supplementary Fig. 1



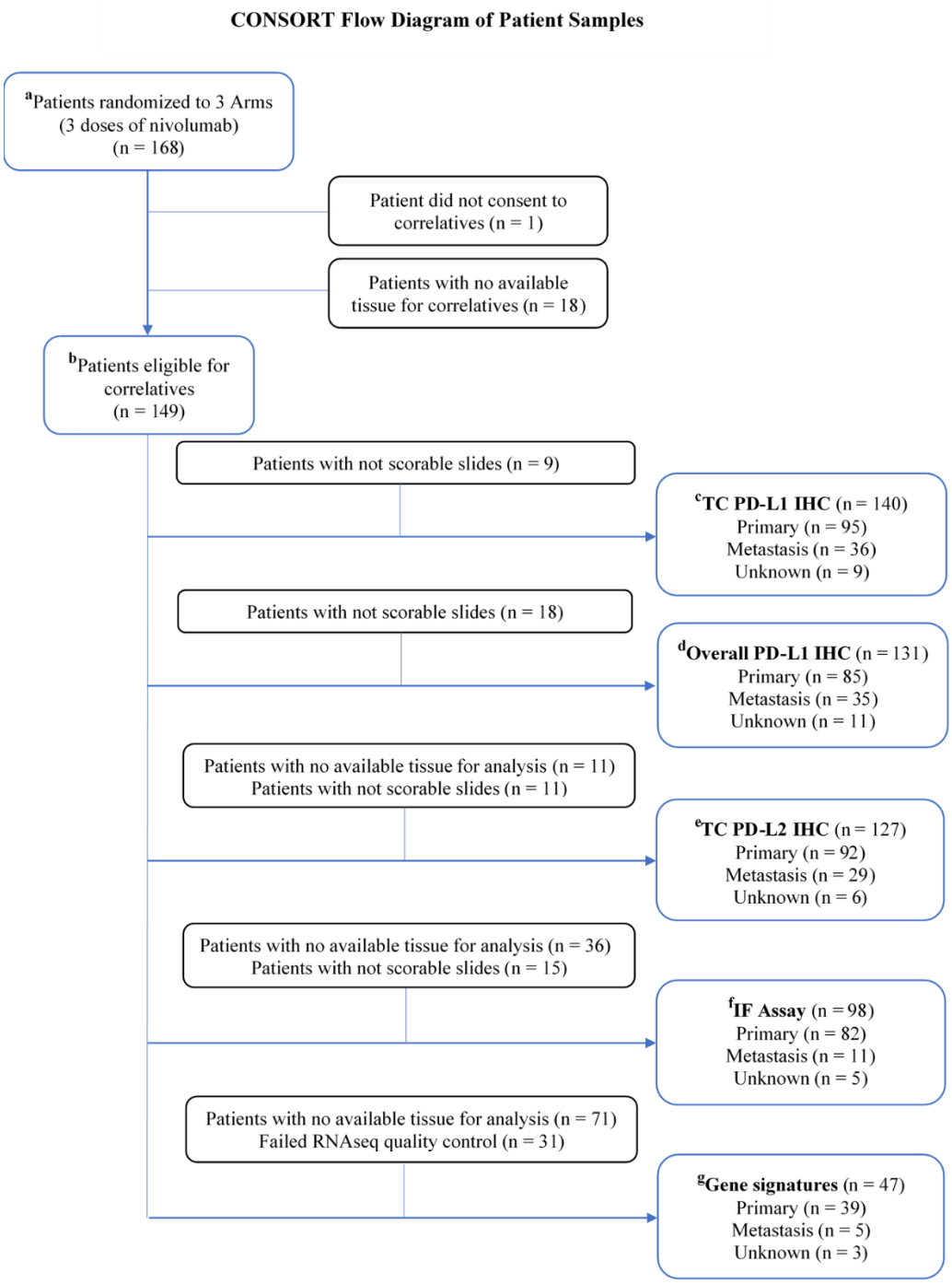
Validation of PD-L2 IHC Assay. Sections of paraffin-embedded 300-19 murine cell line transfected with empty vector (A, C, E) or with vectors coding human PD-L2 (B) or human PD-L1 (D, F) were immunostained with a rabbit monoclonal anti-PD-L2 antibody (clone D7U8C) (A, C, B, D) or a with a mouse monoclonal anti-PD-L1 antibody (clone 9A11) (E, F). Immunoreactivity with the anti-PL2 antibody was detected in PD-L2 transfected 300-19 cells (B) but not in in PD-L1 transfected 300-19 cells (D). IHC against PD-L1 confirmed the presence of PD-L1 in PD-L1 transected cells (F). No immunoreactivity for PD-L1 or PD-L2 was detected in cells transfected with empty vectors (A, C, E). Immunoreactivity for endogenous PD-L2 was detected in the Hodgkin lymphoma cell line HDLM-2 cell line harboring an amplification of the PD-L1/PL-L2 locus (H) while no reactivity was detected in the diffuse large B cell lymphoma OCILy1 cell line known not to express PD-L2 (G) (1).

Supplementary Fig. 2



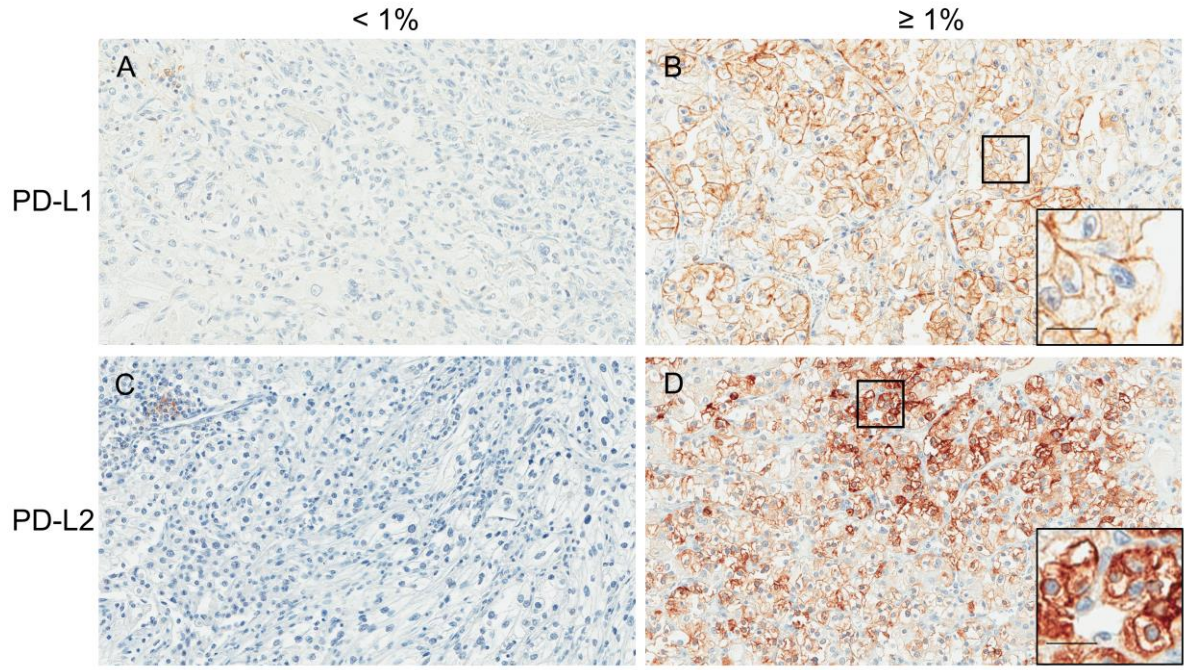
Workflow of Multiplex IF Image Analysis for the Expression of PD-1, TIM-3 and LAG-3 on CD8$^+$ Cells Using the Inform 2.2 Software. Multispectral images (A) were deconvoluted (B), and cell segmentation (C) was then performed. Phenotyping (D) was performed by developing 3 algorithms recognizing cells mono-stained for CD8 or co-stained for CD8 and PD-1 (left panels); cells mono-stained for CD8 or co-stained for CD8 and TIM-3 (middle panels); cells mono-stained for CD8 or co-stained for CD8 and LAG-3 (right panels). A cell was called CD8$^+$ only if it was recognized as CD8$^+$ by all three different algorithms. For each segmented cell, information about the presence or the absence of CD8, PD-1, TIM3 or LAG3 staining was obtained and recorded.

Supplementary Fig. 3



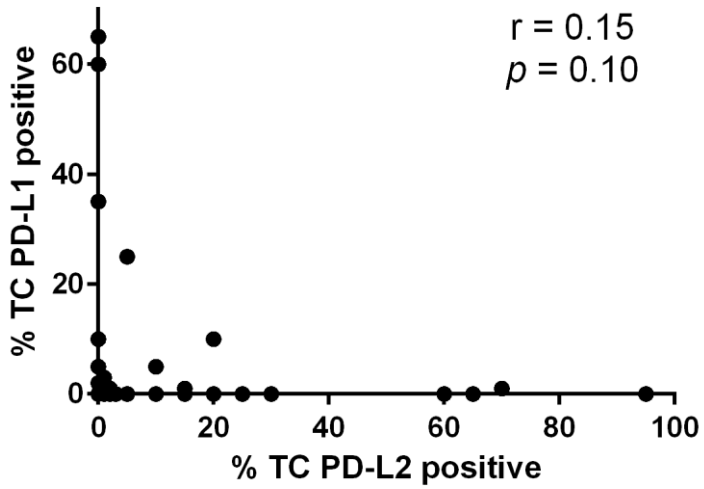
**a,b,c,d,e,f,g: No statistically significant difference in Chi square test of equal proportion of treatment dose arms (minimum p-value was 0.656).*
***Differences in clinical endpoints (ORR & irORR and PFS & irPFS) between patients with missing vs non-missing biomarker measures were assessed for the major and minor node and edges (i.e. a vs b – g, and b vs c – g). The minimum, unadjusted p-value for all combinations of comparisons was 0.166 suggesting no systematic difference in clinical endpoints between cases with and without biomarker measures.*

Supplementary Fig. 4



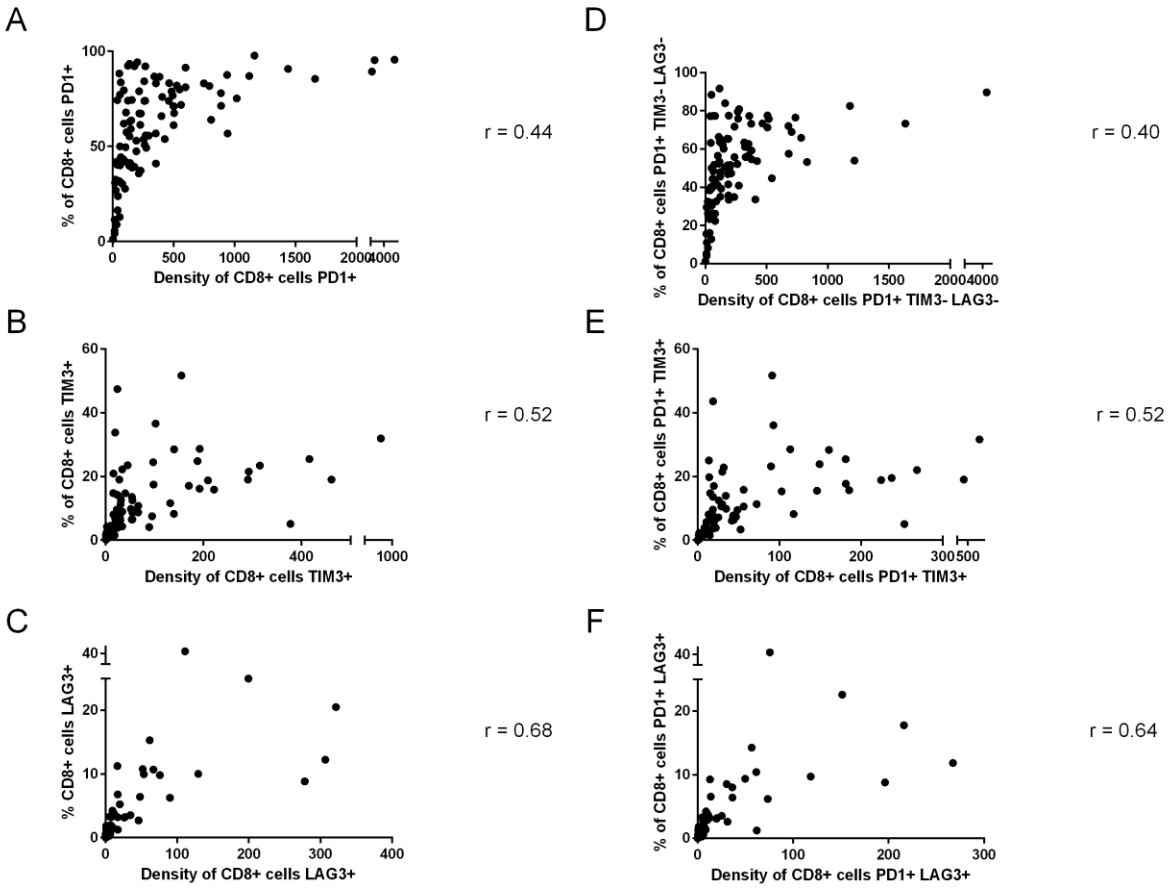
Representative Images of ccRCC Tissue Sections Immunostained for PD-L1 (A, B) or PD-L2 Expression (C, D) from Patients with Positive Tumor Cells <1% (A, C) or ≥ 1% (B, D).
Inset show higher magnification of the selected area (scale bar = 20 µm).

Supplementary Fig. 5



Scatter Plot of the Percentage of Tumor Cells Expressing PD-L1 versus the Percentage of Tumor Cells Expressing PD-L2.

Supplementary Fig. 6



Scatter Plot of the Percentage versus Density of CD8$^+$ Tumor Infiltrating Cells Expressing PD-1, TIM-3 or LAG-3 either Alone (A, B, C) or in Different Combinations (D, E, F).

Reference Supplementary Figures

1. Green MR, Monti S, Rodig SJ, Juszczynski P, Currie T, O'Donnell E, et al. Integrative analysis reveals selective 9p24.1 amplification, increased PD-1 ligand expression, and further induction via JAK2 in nodular sclerosing Hodgkin lymphoma and primary mediastinal large B-cell lymphoma. Blood. 2010 Oct 28;116(17):3268-77.