**Additional file 2**

**Table S1.** Patient demographics and baseline characteristics for analyzed samples.

|  |  |  |
| --- | --- | --- |
|  | Durvalumab-treated cohort  | Non-ICT cohort (n=199) |
| **Training set** **(n=84)** | **Test set (n=79)** | ***p*-valuea** | **All (n=163)** |
| **Sex (%)** |  |  | 0.55 |  |  |
|  **F** | 46.4 | 41.8 |  | 44.2 | 20.1 |
|  **M** | 53.6 | 58.2 |  | 55.8 | 79.9 |
| **Age (%)** |  |  | 0.06 |  |  |
|  **≤35** | 1.2 | 1.3 |  | 1.2 | 0 |
|  **36-45** | 2.4 | 3.8 |  | 3.1 | 6.5 |
|  **46-55** | 8.3 | 25.3 |  | 16.6 | 18.1 |
|  **56-65** | 34.5 | 26.6 |  | 30.7 | 23.6 |
|  **66+** | 53.6 | 43.0 |  | 48.5 | 51.8 |
| **Smoking status (%)** |  |  | 0.90 |  |  |
|  **0-14 pack-years** | 27.4 | 30.4 |  | 28.8 | 12.6 |
|  **15+ pack-years** | 64.3 | 60.8 |  | 62.6 | 82.4 |
|  **Unknown** | 8.3 | 8.9 |  | 8.6 | 5.0 |
| **Histology (%)** |  |  | 0.83 |  |  |
|  **Squamous** | 53.6 | 51.9 |  | 52.8 | 38.7 |
|  **Non-squamous** | 46.4 | 48.1 |  | 47.2 | 57.8 |
|  **Other** | 0.0 | 0.0 |  | 0.0 | 3.5b |
| **T stage (%)** |  |  | 0.88 |  |  |
|  **T0** | 0.0 | 1.3 |  | 0.6 | 0 |
|  **T1** | 11.9 | 12.7 |  | 12.3 | 22.6 |
|  **T2** | 26.2 | 21.5 |  | 23.9 | 43.7 |
|  **T3** | 13.1 | 16.5 |  | 14.7 | 23.6 |
|  **T4** | 19.0 | 22.8 |  | 20.9 | 10.1 |
|  **TX** | 9.5 | 8.9 |  | 9.2 | 0 |
|  **Unknown** | 20.2 | 16.5 |  | 17.8 | 0 |
| **N stage (%)** |  |  | 0.43 |  |  |
|  **N0** | 20.2 | 22.8 |  | 21.5 | 62.3 |
|  **N1** | 14.3 | 5.1 |  | 9.8 | 19.1 |
|  **N2** | 21.4 | 26.6 |  | 23.9 | 18.1 |
|  **N3** | 16.7 | 20.3 |  | 18.4 | 0 |
|  **NX** | 7.1 | 8.9 |  | 8.0 | 0.5 |
|  **Unknown** | 20.2 | 16.5 |  | 17.8 | 0 |
| **M stage (%)** |  |  | 0.51 |  |  |
|  **M0** | 28.6 | 31.6 |  | 30.1 | 100 |
|  **M1** | 48.8 | 54.4 |  | 51.5 | 0 |
|  **MX** | 3.6 | 1.3 |  | 2.5 | 0 |
|  **Unknown** | 19.0 | 12.7 |  | 15.3 | 0 |
| **Cancer staging (%)** |  |  | 0.66 |  |  |
|  **I** | 3.6 | 2.5 |  | 3.1 | 44.2 |
|  **II** | 10.7 | 7.6 |  | 9.2 | 23.6 |
|  **III** | 22.6 | 26.6 |  | 24.5 | 32.2 |
|  **IV** | 61.9 | 60.8 |  | 61.3 | 0 |
|  **Unknown** | 1.2 | 2.5 |  | 1.8 | 0 |
| **Line of therapy (%)** |  |  | 0.93 |  |  |
|  **1** | 23.8 | 25.3 |  | 24.5 | 100 |
|  **2** | 23.8 | 25.3 |  | 24.5 | 0 |
|  **≥3** | 52.4 | 49.4 |  | 50.9 | 0 |
| **PD-L1 status (%)** |  |  | 0.99 |  |  |
|  **<25%** | 41.7 | 40.5 |  | 41.1 |  |
|  **≥25%** | 58.3 | 59.5 |  | 58.9 |  |
| **Response to durvalumab (%)c** |  |  | 0.09 |  |  |
|  **CR** | 1.2 | 0.0 |  | 0.6 |  |
|  **PR** | 17.9 | 19.0 |  | 18.4 |  |
|  **SD** | 25.0 | 43.0 |  | 33.7 |  |
|  **PD** | 41.7 | 24.1 |  | 33.1 |  |
|  **NE** | 14.3 | 13.9 |  | 14.1 |  |

a*p*-value of a Chi-squared test comparing homogeneity between training and test sets. There is no significant difference observed between the training and test set for any of the clinical categories (all *p*-values > 0.05).

bOther NSCLC histology consist of carcinoma, not otherwise specified (n=2), and giant cell carcinoma (n=5).

cResponse rates shown are for patients with suitable tumor specimens included in this analysis.

**Table S2.** Performance of the CD8xPD-L1 signature, its components, and manual PD-L1 TC scoring in the combined set of
durvalumab-treated and non-ICT patients.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Signature (n)** | **Set** | **Prevalence** | **Median OS, months****(95% CI)** | ***p*-value** | **PPV for response(95% CI)** | **Median PFS, months(95% CI)** | ***p*-value**  |
| **CD8xPD-L1 positive (59)** | **Durvalumab**  | 0.362 | **21.0 (17.9–27.9)** | **0.00002** | **0.39 (0.265–0.526)** | 7.3 (4.1–9.2) | 0.00000027 |
| **CD8xPD-L1 negative (104)** | **Durvalumab** | 0.638 | **7.8 (5.4–10.3)** | **0.0769 (0.0338–0.146)** | 1.4 (1.4–2.3) |
| **CD8+ high cell density (74)** | **Durvalumab** | 0.454 | **20.3 (15.5–24.3)** | **0.00013** | **0.284** **(0.185–0.401)** | *5.3* (3.1–7.4) | 0.00000192 |
| **CD8+ low cell density (89)** | **Durvalumab** | 0.546 | **7.6** **(5.1–9.8)** | **0.112** **(0.0552–0.197)** | 1.7 (1.4–2.6) |
| **PD-L1+ high cell density (55)** | **Durvalumab** | 0.337 | **20.3 (14.0–27.9)** | **0.0064** | **0.382 (0.254–0.523)** | 5.3 (2.8–7.4) | 0.00231 |
| **PD-L1+ low cell density (108)** | **Durvalumab** | 0.663 | **9.3 (6.5–13.1)** | **0.0926 (0.0453–0.164)** | 1.7 (1.4–2.8) |
| **PD-L1 TC ≥25% (96)** | **Durvalumab** | 0.589 | **17.9 (10.3–24.2)** | **0.0082** | **0.281 (0.194–0.382)** | 3.6 (2.6–5.3) | 0.00411 |
| **PD-L1 TC <25% (67)** | **Durvalumab** | 0.411 | **7.8 (6.0–11.1)** | **0.0597 (0.0165–0.146)** | 1.4 (1.3–2.6) |
| **CD8xPD-L1 positive (72)** | **Non-IO** | 0.362 | **52** **(37–89)** | **0.58** | **NA** **(0–1)** | NA | NA |
| **CD8xPD-L1 negative (127)** | **Non-IO** | 0.638 | **56** **(42–69)** | **NA** **(0–1)** | NA |
| **CD8+ high cell density (135)** | **Non-IO** | 0.678 | **67** **(50–92)** | **0.00085** | **NA** **(0–1)** | NA | NA |
| **CD8+ low cell density (64)** | **Non-IO** | 0.322 | **39.5****(21– 56)** | **NA****(0–1)** | NA |
| **PD-L1+ high cell density (54)** | **Non-IO** | 0.271 | **43.5** **(31–59)** | **0.25** | **NA** **(0–1)** | NA | NA |
| **PD-L1+ low cell density (145)** | **Non-IO** | 0.729 | **60** **(47–81)** | **NA** **(0–1)** | NA |
| **PD-L1 TC ≥25% (30)** | **Non-IO** | 0.152 | **30** **(15–50)** | **0.004** | **NA** **(0–1)** | NA | NA |
| **PD-L1 TC <25% (168)** | **Non-IO** | 0.848 | **61** **(51–82)** | **NA** **(0–1)** | NA |

**Table S3.** Multiparametric Cox analysis of signatures in the entire data set including additional PD-L1 readouts.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Significant fixed covariate** | **Significance (fixed covariate)** | **Added covariate** | **Significance (added covariate)** | **Significance (Cox model)** |
| **Liver metastasis/line of therapy** | ++/+ | **None** | N/A | + |
| **LM** | +++ | **CD8xPD-L1 signature** | ++++ | ++++ |
| **LM** | + | **CD8+ cell density** | ++ | ++ |
| **LM** | ++ | **PD-L1 TC expression** | + | + |
| **LM** | +++ | **PD-L1+ cell density**  | + | ++ |
| **LM** | +++ | **% of PD-L1+ pixelsa** | + | ++ |
| **LM** | +++ | **% of PD-L1+ cellsb** | + | ++ |

Fixed covariates at baseline were histology, smoking status, age, gender, liver metastasis, tumor stage and line of therapy. + ≤ 0.05; ++ ≤ 0.005; +++ ≤ 0.0005; ++++ ≤ 0.00005.
aPercentage of PD-L1+ pixels: Ratio of PD-L1+ pixels to total pixels in tissue
bPercentage of PD-L1+ cells: Ratio of PD-L1+ cells to all cells