PD-L1 IMMUNOHISTOCHEMISTRY OPTIONS

<table>
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<tr>
<th>TEST CODE</th>
<th>CLONE</th>
<th>SOURCE</th>
<th>TUMOR</th>
<th>ORIGINAL FDA-APPROVED DRUG</th>
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<tr>
<td>PDL12</td>
<td>22C3</td>
<td>Dako</td>
<td>non-small-cell lung cancer (NSCLC)</td>
<td>Companion diagnostic assay – KEYTRUDA® (pembrolizumab, Merck)</td>
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<td>recurrent or advanced gastric or gastroesophageal junction adenocarcinoma</td>
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<tr>
<td>PDL1</td>
<td>SP263</td>
<td>Ventana</td>
<td>previously treated NSCLC</td>
<td>Complementary diagnostic assay – OPDIVO® (nivolumab, Bristol-Myers Squibb)</td>
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<td>advanced or metastatic urothelial carcinoma</td>
<td>Complementary diagnostic assay – IMFINZI® (durvalumab, AstraZeneca)</td>
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<tr>
<td>PDL1S</td>
<td>SP142</td>
<td>Ventana</td>
<td>metastatic urothelial cancer</td>
<td>Complementary diagnostic assay – TECENTRIQ® (atezolizumab, Genentech)</td>
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**CLINICAL INFORMATION**

Programmed cell death-ligand 1 (PD-L1) is a transmembrane protein involved in the regulation of cell-mediated immune responses through interaction with the programmed cell death-1 (PD-1) protein. PD-L1 has been identified as both a prognostic and theranostic marker in a variety of neoplasms. Overexpression of PD-L1 has been observed in carcinomas of the urinary bladder, lung, thymus, colon, pancreas, ovary, breast, kidney and in melanoma, malignant mesothelioma, and glioblastoma.

**INTERPRETATION**

For all PD-L1 clones, results will be reported as percent-positive tumor cells and whether >5% of immune cells express PD-L1. For gastric and GE junction adenocarcinomas using the 22C3 clone, results will be reported as a combined positive score (CPS). If additional interpretation or analysis is needed, request test PATHC/Pathology Consultation.

**SPECIMEN REQUIRED**

The specimen needs to be a formalin-fixed, paraffin-embedded tissue block, or it must be 3 unstained slides with 4-microns formalin-fixed, paraffin-embedded tissue. The use of cell blocks and decalcified specimens has not been validated.

Adenocarcinoma stained with PD-L1 clone 22C3.
FREQUENTLY ASKED QUESTIONS

Q Why does Mayo Clinic Laboratories (MCL) offer multiple tests for PD-L1?
A MCL currently offers three clones of the PD-L1 antibody for IHC staining: clone 22C3, clone SP263, and clone SP142. The 22C3 clone is a Dako antibody run on the Ventana platform; the SP263 and SP142 clones are Ventana antibodies run on the Ventana platform. The reason for offering three different clones is that each clone is associated with a different drug and can have a unique staining pattern and interpretation guidelines.

Q When should the 22C3 clone be used?
A Pembrolizumab is an FDA-approved anti-PD-1 drug used as a first line of treatment for non-small-cell lung carcinoma (NSCLC). The approval for pembrolizumab is linked to a companion diagnostic assay. To be eligible for this therapy, patients must have their tissue tested with the 22C3 antibody clone and must have positive staining by 50% or more of the tumor cells. More recently, pembrolizumab has been approved for use in advanced gastric or gastroesophageal junction carcinomas. The same 22C3 antibody is required, however, interpretation guidelines are different. For these tumors, PD-L1 expression is assessed as a combined positive score, meaning that all cells (tumor cells, lymphocytes, macrophages) positive for PD-L1 are compared to the total number of tumor cells. The tumor tissue must have a CPS ≥1 in order to be eligible for therapy.

Q When should the SP142 and SP263 clones be used?
A Other therapies have complementary tests associated with them where the testing is highly recommended but not required in order to prescribe therapy. SP263 is used as a complementary assay in NSCLC for nivolumab or in metastatic urothelial carcinoma for durvalumab. SP142 is used as a complementary assay in both metastatic urothelial carcinoma and NSCLC for atezolizumab.

Q Are the three clones all equivalent?
A Generally, clones 22C3 and SP263 have similar staining patterns. Clone SP142 often stains fewer cells—both tumor cells and immune cells. However, because of the different therapies and tumor types associated with each clone, none of the clones should be used interchangeably.

Q What is unique or challenging about the interpretation of PD-L1?
A Both tumor cells and immune cells can express PD-L1. In tumor types that have many macrophages, such as NSCLC, occasionally, it can be difficult to differentiate between the tumor cells and the immune cells. Also, PD-L1 expression can be very heterogeneous within a tumor, meaning that expression in a single biopsy may not be reflective of the entire tumor. There is also heterogeneity in the expression of PD-L1 between the primary tumor and metastatic tumor, between the primary tumor and recurrent tumor, and among multiple primary tumors.