COLLEGE of AMERICAN PATHOLOGISTS

ARCHIVES
of Pathology & Laboratory Medicine

NEW ARTICLE

This article was posted on the *Archives* Web site as a New Article. New Articles have been peer reviewed, copyedited, and reviewed by the authors. Additional changes or corrections may appear in these articles when they appear in a future print issue of the *Archives*. New Articles are citable by using the Digital Object Identifier (DOI), a unique number given to every article. The DOI will typically appear at the end of the abstract.

The DOI for this manuscript is doi: 10.5858/arpa.2023-0536-CP

The print version of this manuscript will replace the New Article version at the above DOI once it is available.

Programmed Death Ligand-1 and Tumor Mutation Burden Testing of Patients With Lung Cancer for Selection of Immune Checkpoint Inhibitor Therapies

Guideline From the College of American Pathologists, Association for Molecular Pathology, International Association for the Study of Lung Cancer, Pulmonary Pathology Society, and LUNGevity Foundation

Lynette M. Sholl, MD; Mark Awad, MD, PhD; Upal Basu Roy, PhD, MS, MPH; Mary Beth Beasley, MD; Richard Walter Cartun, PhD, MS; David M. Hwang, MD, PhD; Gregory Kalemkerian, MD; Fernando Lopez-Rios, MD, PhD; Mari Mino-Kenudson, MD; Ajit Paintal, MD; Kearin Reid, MLS(ASCP), MLIS; Lauren Ritterhouse, MD, PhD; Lesley A. Souter, PhD; Paul E. Swanson, MD; Christina B. Ventura, MPH, MT(ASCP); Larissa V. Furtado, MD

• ***Context.***—**Rapid advancements in the understanding and manipulation of tumor-immune interactions have led to the approval of immune therapies for patients with non–small cell lung cancer. Certain immune checkpoint inhibitor therapies require the use of companion diagnostics, but methodologic variability has led to uncertainty around test selection and implementation in practice.**

Objective.—**To develop evidence-based guideline recommendations for the testing of immunotherapy/immunomodulatory biomarkers, including programmed death ligand-1 (PD-L1) and tumor mutation burden (TMB), in patients with lung cancer.**

Design.—**The College of American Pathologists convened a panel of experts in non–small cell lung cancer and biomarker testing to develop evidence-based recommendations in accordance with the standards for trustworthy clinical practice guidelines established by the National Academy of Medicine. A systematic literature review was conducted to address 8 key questions. Using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach, recommendations were created from the available evidence, certainty of that evidence, and key judgments as defined in the GRADE Evidence to Decision framework.**

Results.—**Six recommendation statements were developed.**

Conclusions.—**This guideline summarizes the current understanding and hurdles associated with the use of PD-L1 expression and TMB testing for immune checkpoint inhibitor therapy selection in patients with advanced non–small cell lung cancer and presents evidence-based recommendations for PD-L1 and TMB testing in the clinical setting.**

(*Arch Pathol Lab Med*. doi: 10.5858/arpa.2023-0536-CP)

The advent of immune modulatory therapy has led to a radical shift in the treatment paradigm for patients with a wide range of cancers. The clinical impact of these therapies, especially those targeting immune "checkpoints," has been particularly profound for patients with non–small cell lung carcinoma (NSCLC). Clinical trials have demonstrated that drugs that block programmed death receptor-1 (PD-1, encoded by *PDCD1*) and programmed death ligand-1 (PD-L1, also known as B7H1, encoded by *CD274*) lead to significant improvements in both response and survival relative to conventional cytotoxic chemotherapy for patients with advanced-stage NSCLC. These trials have identified PD-L1 protein expression, based on immunohistochemistry (IHC), as a biomarker for improved benefit following anti–PD-1/PD-L1 therapies and combination therapies (hereafter referred to as immune

Accepted for publication February 29, 2024.

Supplemental digital content is available for this article. See text for hyperlink.

From the Department of Pathology, Brigham and Women's Hospital, Boston, Massachusetts (Sholl); Lowe Center for Thoracic Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts (Awad); Translational Science Research Program, LUNGevity Foundation, Chicago, Illinois (Basu Roy); the Department of Anatomic Pathology and Clinical Pathology, Mt. Sinai Medical Center, New York, New York (Beasley); the Department of Anatomic Pathology, Hartford Hospital, Hartford, Connecticut (Cartun); the Department of Laboratory Medicine & Pathobiology, Sunnybrook Health Science Centre, Toronto, Ontario, Canada (Hwang); the Department of Medical Oncology and Internal Medicine, University of Michigan Health, Ann Arbor (Kalemkerian); Pathology Department, Hospital Universitario 12 de Octubre, Madrid, Spain (Lopez-Rios); the Department of Pathology, Massachusetts General Hospital, Boston (Mino-Kenudson); the Department of Pathology, NorthShore University Health System, Evanston, Illinois (Paintal); Governance (Reid) and the Pathology and Laboratory Quality Center for Evidence-based Guidelines (Ventura), College of American Pathologists, Northfield, Illinois; the Department of Pathology, Foundation Medicine, Cambridge, Massachusetts (Ritterhouse); Methodology Consultant, Hamilton, Ontario, Canada (Souter); the Department of Laboratory Medicine and Pathology, University of Washington Medical Center, Seattle (Swanson); and the Department of Pathology, St. Jude Children's Research Hospital, Memphis, Tennessee (Furtado). Cartun is now retired. Sholl and Furtado are the Guideline Expert Panel Co-chairs.

Authors' disclosures of potential conflicts of interest and author contributions are found in the Appendix at the end of this article.

Corresponding author: Lynette M. Sholl, MD, Department of Pathology, Brigham and Women's Hospital, 75 Francis St, Boston MA 02115 (email: lmsholl@bwh.harvard.edu).

checkpoint inhibitors, or ICIs). The biomarker testing space has been complicated, however, by the proliferation of PD-L1 assays and scoring criteria that have evolved with individual therapies and often for different tumor types, some of which have received companion diagnostic (CDx) approvals by regulatory agencies such as the US Food and Drug Administration (FDA) and the Health Products and Food Branch of Health Canada.[1] At the same time, for reasons of cost and access, PD-L1 IHC antibodies and assays developed outside of the scope of randomized controlled trials (RCTs) have garnered widespread use,[2] thus leading to confusion on the part of pathologists and clinicians about the best approach to biomarker testing to select patients for ICI therapy. In contrast to most genomic biomarkers (eg, *anaplastic lymphoma kinase* [*ALK*], *epidermal growth factor receptor* [*EGFR*]), which tend to represent relatively stable and binary data points in a patient's tumor profile in treatment-naive patients, PD-L1 expression is dynamic and heterogeneous, complicating the choice of sample for testing.[3–5]

At the same time, there is ongoing interest in genomic biomarkers in immunotherapy—in particular, tumor mutation burden (TMB)—that may be used in conjunction with or independent of PD-L1 status. Indeed, high TMB has been approved by the FDA as a cancer-type agnostic biomarker for the anti–PD-1 monoclonal antibody pembrolizumab, thus elevating this test to clinical relevance, despite numerous challenges relating to access, technical reproducibility, and biological relevance of the proposed cutoff.

The guideline's primary goal is to develop evidence-based recommendations for the testing of immunotherapy biomarkers, including PD-L1 and TMB, in patients with NSCLC. Several ICI-based therapies have been approved by regulatory agencies globally in the first and second lines of therapy for patients with NSCLC. Companion diagnostics are required for certain therapies, but for reasons of cost and access to necessary reagents and equipment, variable methodology exists in practice. As a result, questions regarding assay interchangeability persist.

METHODS

This evidence-based guideline was developed following the standards by the National Academy of Medicine.[6] A detailed description of the panel composition, conflict of interest (COI) policy, and systematic review methods used to create this guideline can be found in the online Evidence-Based Guidelines Development Methodology Manual (Methodology Manual).[7]

Guideline Panel

The College of American Pathologists (CAP), in collaboration with the American Society of Clinical Oncology (ASCO), Association for Molecular Pathology (AMP), International Association for the Study of Lung Cancer (IASLC), Pulmonary Pathology Society (PPS), and the patient advocacy organization LUNGevity Foundation, convened a multidisciplinary expert and advisory panel to develop the guideline and approved the appointment of the members. Members included practicing pathologists, biomedical scientists, oncologists, patient advocates, and a guideline methodologist. Panel members were selected to represent diverse laboratory environments and geographic locations to assure that multiple perspectives would be represented. The roles of each panel are described in the Methodology Manual. Detailed information about the panel composition can be found in the supplemental digital content (SDC). The expert panel (EP) met via teleconference and 1 in-person meeting, using a modified Delphi method to come to agreements about the guideline scope and recommendations. Work was also conducted via email communication.

Conflict of Interest

In accordance with the CAP COI policy, members of the EP disclosed all financial interests from 2 years prior to appointment through the development of the guideline. Complete disclosures of the EP members are listed in the Appendix. A detailed description of the policy is included in the Methodology Manual.

The majority of the EP (10 of 12 members) was assessed as having no relevant COI. Disclosures of interest judged by the oversight group to be manageable conflicts are as follows. Mark Awad: consulting fees or advisory board, Affini-T Therapeutics, Inc (Watertown, Massachusetts), AstraZeneca (Wilmington, Delaware), Bristol-Myers Squibb (Princeton, New Jersey), EMD Serono (Rockland, Massachusetts), Genentech (South San Francisco, California), Gritstone bio (Emeryville, California), Foundation Medicine (Boston, Massachusetts), Instil Bio (Dallas, Texas), Janssen Oncology (Titusville, New Jersey), Merck (Rahway, New Jersey), Mirati Therapeutics Inc (San Diego, California), Novartis (East Hanover, New Jersey), Pfizer (New York, New York), Regeneron Pharmaceuticals (Tarrytown, New York); research grants, AstraZeneca (Wilmington, Delaware), Bristol-Myers Squibb (Princeton, New Jersey). Lauren Ritterhouse: employment and stock options, Foundation Medicine (Boston, Massachusetts).

The CAP provided funding for the administration of the project; no industry funds were used in the development of the guideline. All panel members volunteered their time and were not compensated for their involvement, except for the contracted methodologist.

Guideline Objectives

The panel addressed the following overarching questions, "Does PD-1/PD-L1 status and TMB improve clinical outcomes in patients with NSCLC who are being considered for ICI therapy?" and "What testing and specimen requirements provide accurate test results for PD-1/PD-L1 and TMB?" This led to the following key questions:

1. In patients with advanced-stage NSCLC who are being considered for ICI therapy, does PD-L1 and TMB testing improve treatment response rates and survival rates?
2. When selecting patients for anti–PD-1 and anti–PD-L1 therapy, does testing of different specimen types provide concordant clinical outcomes?
3. Does the use of ICI therapy in patients with advanced NSCLC with targetable *ALK*, *EGFR*, *ROS proto-oncogene 1* (*ROS1*), or *B-Raf proto-oncogene* (*BRAF*) molecular alterations affect their long-term clinical outcomes?
4. When selecting patients for anti–PD-1 and anti–PD-L1 therapy, does TMB testing have the analytic validity to identify a complementary population who will benefit from therapy?
5. In patients with NSCLC with more than 1 available sample, do multiple samples provide concordant PD-L1 and TMB testing results and downstream clinical outcomes?
6. Does clinical validity of PD-L1 testing differ by levels of PD-L1 expression in tumor or immune cells?
7. How reproducible are PD-L1 tumor cell scores and immune cell scores across specimen types?
8. Do the available PD-L1 assays provide concordant expression profiles when evaluating the same sample, and which IHC expression cutoff provides the most reproducible expression categorization across the assays?

See Supplemental Table 1 in the SDC for more details.

Literature Search and Collection

A comprehensive literature search for relevant evidence was completed by a medical librarian. The search strategy was first constructed in Ovid MEDLINE (Wolters Kluwer Health, Inc), using controlled vocabulary and keywords agreed upon by the EP to reflect the population, intervention, comparator, and outcome elements, then translated into Embase (Elsevier) and Cochrane Library (John Wiley & Sons, Inc). Major concepts included NSCLC, PD-L1/PD-1, TMB, and

Table 1. Certainty of Evidence[a]	
Designation	**Description**
High	There is high confidence that available evidence reflects true effect. Further research is very unlikely to change the confidence in the estimate of effect.
Moderate	There is moderate confidence that available evidence reflects true effect. Further research is likely to have an important impact on the confidence in estimate of effect and may change the estimate.
Low	There is limited confidence in the estimate of effect. The true effect may be substantially different from the estimate of the effect.
Very Low	There is very little confidence in the estimate of effect. The true effect is likely to be substantially different from the estimate of effect. Any estimate of effect is very uncertain.

[a] Data derived from Grading of Recommendations Assessment, Development and Evaluation (GRADE) Working Group materials.[8]

laboratory testing. Limits were set to reflect the protocol inclusion/exclusion criteria, including (1) the publication date range of January 1, 2010, through the date each search was run; (2) language filters to capture only full-text articles available in English owing to time and financial constraints; and (3) publication limits to exclude letters, editorials, commentaries, and case studies. The Cochrane filter was applied to exclude animal studies. The database literature searches were initially run on October 16, 2019, and rerun on April 7, 2021, and May 13, 2022, to capture studies published since the initial searches were run. Supplemental searches to complement the database references were completed, and EP members were polled for relevant unpublished data at the onset of the project. See the SDC Supplemental Figures 1 and 2 for more information, including specific search strategies, supplemental search sources used, dates of search activity, and the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) table that details the systematic review process.

Inclusion and Exclusion Criteria

Studies were selected for inclusion in the systematic review of evidence if they met the following criteria: (1) peer-reviewed articles published since January 1, 2010; (2) study population consisted of adult patients with early- or advanced-stage NSCLC either receiving or undergoing selection for checkpoint inhibitor therapy; (3) study compared, prospectively or retrospectively, laboratory testing methodologies for PD-L1; and (4) study included measurable data such as diagnostic test characteristics, accuracy of PD-L1 expression, survival outcomes, or treatment response.

Articles were excluded from the systematic review if (1) they were published before 2010; (2) they were editorials, letters, commentaries, and invited opinions; (3) the full-text article was not available in English; and (4) they did not address at least 1 key question or the outcomes of interest.

Assessing the Strength of Recommendations

Development of recommendations required the EP to review the identified evidence and make a series of key judgments, using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach.[8] See Supplemental Table 2 for the definitions of strength of recommendation. Supplemental Table 3 in the SDC provides a summary of the key judgments the panel considered, including the benefits and harms of each guideline statement using the GRADE Evidence to Decision (EtD) framework.[9]

Assessing Quality and Risk of Bias

Each study received a risk of bias assessment, and each recommendation was assigned an aggregate assessment of the certainty of evidence (Table 1). Refer to the SDC for definitions of the certainty of evidence (Supplemental Table 4) and for the individual study and aggregate risk of bias assessment (Supplemental Tables 5 through 9).

RESULTS

A total of 3089 studies met the eligibility requirements for screening. Based on review of these titles and abstracts, 356 articles met the inclusion criteria and continued to full-text review. A total of 121 articles were included for data extraction and qualitative analysis. Excluded articles were available as discussion or background references. Additional information is available in the SDC, including a PRISMA table outlining the details of the systematic review. Refer to the write-up for each recommendation for specific details about supporting evidence.

The EP convened by teleconferences and 1 face-to-face meeting to develop the scope, draft recommendations, review and respond to solicited feedback, and assess the certainty of evidence that supports the final recommendations presented herein. A modified Delphi technique was used for consensus decision-making to encourage balanced input and participation among members. An open comment period was posted on the CAP website (www.cap.org) from March 31 to April 23, 2021, during which the draft recommendation statements were available for public feedback. Refer to the SDC for more details.

The EP approved the final recommendations with a supermajority vote (ie, at least 75% of panel members in agreement). An independent review panel, masked to the EP and vetted through the COI process, recommended approval by the CAP Council on Scientific Affairs. The manuscript was also reviewed and approved by the collaborating associations. The final recommendations are summarized in Table 2.

Recommendation Statements

1. In patients with advanced non–small cell lung cancer, pathologists should use a validated PD-L1 immunohistochemistry expression assay, in conjunction with other targetable genomic biomarker assays where appropriate, to optimize selection for treatment with immune checkpoint inhibitors.

(Strength of Recommendation: Strong; Certainty of Evidence: Moderate)

The evidence for this statement included a total of 34 studies[10–43] that evaluated overall survival (OS) rates and response rate (RR) following treatment with various immunotherapy agents in tumors with known PD-L1 expression status. The certainty of evidence was moderate for both outcomes of interest. From the available evidence, EP members concluded that OS and RR with immunotherapy were correlated with PD-L1 expression status. After discussions, EP members defined the benefits of PD-L1 expression detection using a validated IHC assay as moderate and the harms of this testing as small, and concluded that the benefits thus

Table 2. Summary of Guideline Statements	
Guideline Statement	**Strength of Recommendation**
1. In patients with advanced NSCLC, pathologists should use a validated PD-L1 IHC expression assay, in conjunction with other targetable genomic biomarker assays where appropriate, to optimize selection for treatment with ICIs.	Strong recommendation
2. Pathologists should ensure appropriate validation has been performed on all specimen types and fixatives. *Note:* Specific validation requirements are out of the scope of this guideline, and laboratories should refer to the Principles of Analytic Validation of Immunohistochemical Assays Guideline[57] for details on how to validate IHC specimens.	Conditional recommendation
3. When feasible, pathologists should use clinically validated PD-L1 IHC assays as intended.	Conditional recommendation
4. Pathologists who choose to use LDTs for PD-L1 expression should validate according to the requirements of their accrediting body.	Strong recommendation
5. Pathologists should report PD-L1 IHC results using a percentage expression score.	Conditional recommendation
6. Clinicians should not use tumor mutation burden alone to select patients with advanced NSCLC for ICIs, based on insufficient evidence in this population.	Conditional recommendation

Abbreviations: ICIs, immune checkpoint inhibitors; IHC, immunohistochemistry; LDTs, laboratory-developed tests; NSCLC, non–small cell lung cancer; PD-L1, programmed death ligand-1.

outweighed the harms. It is expected that this guidance will be acceptable to key stakeholders and feasible to implement. Refer to Supplemental Tables 5 through 8 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement. Supplemental Table 3 summarizes the EtD framework.

Published studies have demonstrated a statistically significant correlation between the presence and extent of PD-L1 expression in tumor tissue samples or tumor proportion score (TPS) and patient response and survival following immunotherapy with PD-1 and PD-L1 inhibitors given alone or in combination with chemotherapy and/or cytotoxic T-lymphocyte–associated protein-4 (CTLA-4) inhibitors (Table 3). To date, these associations have been most widely demonstrated in the subset of patients with stage IV metastatic nonsquamous (mainly adenocarcinoma) and squamous cell NSCLC. PD-L1 ICI therapy with atezolizumab has also shown a disease-free survival benefit in the adjuvant setting for patients with PD-L1–positive (TPS $\geq$1%), resected NSCLC.[44] Similarly, PD-1 ICI therapy with pembrolizumab has been approved in the adjuvant setting for patients with resected IB-IIIA NSCLC, regardless of PD-L1 expression status. PD-1 therapy with nivolumab in combination with chemotherapy has also been approved in the neoadjuvant setting for patients with surgically resectable NSCLC irrespective of PD-L1 status.[45] However, owing to the timing of the literature review and guideline drafting (which occurred primarily before the trials supporting these approvals were published), data related to the use of PD-L1 and related biomarkers in early-stage NSCLC are out of scope for this guideline.

Although there is an association between PD-L1 expression and response for all lines of therapy in patients with advanced disease, these associations are strongest in the first-line setting. From phase-3 RCTs comparing PD-1/PD-L1 inhibitors to chemotherapy as first-line treatment in patients with advanced NSCLC, the FDA approved monotherapy with pembrolizumab, atezolizumab, or cemiplimab in patients whose tumors showed a PD-L1 TPS of 50% or more, and combination nivolumab + ipilimumab in patients with tumors expressing PD-L1 at TPS of 1% or more.[9,45] Of note, the first-line atezolizumab approval also included patients with tumors harboring PD-L1–positive tumor-infiltrating immune cells covering 10% or more of the tumor area. A subsequent phase-3 RCT in patients with advanced NSCLC comparing first-line pembrolizumab monotherapy to chemotherapy showed a statistically significant survival benefit with pembrolizumab in the subgroup of patients with a PD-L1 TPS of 1% or more; however, this benefit was largely driven by superior outcomes in those with a TPS of 50% or more.[46] Nevertheless, the FDA expanded the indication for first-line pembrolizumab monotherapy to include people with tumors with a PD-L1 TPS of 1% or more. However, this strategy has not been embraced globally, with other international regulatory agencies adhering to the threshold of 50% or more.[47]

Around the world, several other immunotherapy and chemoimmunotherapy combinations have also been approved in the first-line setting for patients with advanced NSCLC, based on clinical trials that have used a specific PD-L1 IHC assay with a scoring cut point to define PD-L1 positivity (Table 3). Some of these trials required certain levels of PD-L1 TPS for enrollment, but many did not, and while the benefits of chemotherapy-ICI combinations have been documented in patients across the PD-L1 TPS spectrum, most studies show improved outcomes with higher levels of PD-L1 expression. The EP recognizes that PD-L1 expression is not an absolute predictive biomarker, with responses and benefit seen in some patients with no visible expression in their tumor sample and lack of benefit in some patients despite high PD-L1 TPS. Nevertheless, considering the plethora of treatment options now available for patients with advanced NSCLC, PD-L1 TPS can be a useful biomarker to inform treatment decision-making and balance potential benefits and risks for individual patients.

Multiple clinical trials of ICI therapy for NSCLC have demonstrated a relative lack of benefit for those patients with driver alterations in the *EGFR* or *ALK* genes.[12] Subsequently, many, but not all, trials of ICI or chemotherapy-ICI in NSCLC have excluded patients with *EGFR* and *ALK* alterations. Furthermore, prospective studies of treatment with concurrent durvalumab plus the EGFR tyrosine kinase inhibitor (TKI) osimertinib in patients with *EGFR*-mutant NSCLC were halted owing to unacceptable levels of interstitial pneumonitis (IP). An analysis of the FDA Adverse Event Reporting System found that the combination of nivolumab plus an EGFR TKI led to a significantly increased risk for development of IP relative to EGFR TKI therapy alone, with the IP events largely restricted to the Japanese population.[48] However, a US-based single-center retrospective analysis also found that 15% of patients who received pembrolizumab followed by osimertinib had severe immune-related adverse events.[49] In

Table 3. Reported Overall Survival and Response Rates for First-Line Immune Checkpoint Inhibitors in Identified Randomized Controlled Trials

RCT	FDA Approval, Y/N	Treatment Arms	PD-L1 IHC Clone	Overall Response Rate	Overall Survival
KEYNOTE-024, Reck et al,[39] 2021	Y	Pembrolizumab, n = 154 Chemotherapy, n = 151	22C3	TPS ≥50%: 46.1%; 95% CI, 38.1%–54.3%	TPS ≥50%: HR, 0.50; 95% CI, 0.39–0.65
KEYNOTE-042, Mok et al,[17] 2019	Y	Pembrolizumab, n = 637 Chemotherapy, n = 637	22C3	TPS ≥1%: 27%; 95% CI, 24%–31% TPS ≥50%: 39%; 95% CI, 34%–45%	Not reported
KEYNOTE-189, Gadgeel et al,[170] 2020	Y	Pembrolizumab + chemotherapy, n = 410 Placebo, n = 206	22C3	TPS 1%–49%: 49.2%; 95% CI, 40.3%–58.2% TPS ≥50%: 62.1%; 95% CI, 53.3%–70.4%	TPS 1%–49%: HR, 0.62; 95% CI, 0.42–0.92 TPS ≥50%: HR, 0.59; 95% CI, 0.39–0.88
KEYNOTE-407, Paz-Ares et al,[171] 2020	Y	Pembrolizumab + chemotherapy, n = 278 Placebo + chemotherapy, n = 281	22C3	TPS ≥1%: 59.1%; 95% CI, 51.4%–66.4%	TPS ≥1%: HR, 0.67; 95% CI, 0.51–0.87
KEYNOTE-010, Herbst et al,[41] 2021	Y	Pembrolizumab, n = 690 Chemotherapy, n = 343	22C3	TPS 1%–49%: 21.2%; 95% CI, 18.2%–24.4% TPS ≥50%: 33.1%; 95% CI, 27.7%–38.8%	TPS 1%–49%: HR, 0.70; 95% CI, 0.61–0.80; $P < .0001$ TPS ≥50%: HR, 0.55; 95% CI, 0.44–0.69; $P < .0001$
CheckMate 017, Brahmer et al,[13] 2015	Y	Nivolumab, n = 135 Chemotherapy, n = 137	28-8	TPS ≥1%[a]: 17% TPS ≥50%: 19%	TPS ≥1%[a]: HR, 0.69; 95% CI, 0.45–1.1 TPS ≥50%: HR, 0.50; 95% CI, 0.28–0.89
CheckMate 026, Carbone et al,[172] 2017	Y	Nivolumab, n = 271 Chemotherapy, n = 270	28-8	TPS ≥5%: OR, 0.70; 95% CI, 0.46–1.06	TPS ≥5%: HR, 1.02; 95% CI, 0.80–1.30
CheckMate 227, Hellmann et al,[16] 2019	Y	Nivolumab + ipilimumab, n = 396 Chemotherapy, n = 396	28-8	TPS ≥1%[a]: 35.9%; 95% CI, 31.1%–40.8% TPS ≥50%: 44.4%; 95% CI, 37.5%–51.1%	TPS ≥1%[a]: HR, 0.79; 95% CI, 0.65–0.96; $P = .007$ TPS ≥50%: HR, 0.70; 95% CI, 0.55–0.90
IMpower110, Herbst et al,[119] 2020	Y	Atezolizumab, n = 277 Chemotherapy, n = 137	SP142	Not reported	TC2/3 or IC2/3: HR, 0.72; 95% CI, 0.52–0.99; $P = .04$ TC3/IC3[a]: HR, 0.59; 95% CI, 0.40–0.87; $P = .01$
IMpower150, Socinski et al,[173] 2021	Y	Atezolizumab + chemotherapy, n = 402 Chemotherapy, n = 400 Atezolizumab + chemotherapy, n = 299 Chemotherapy, n = 279	SP142 SP263	Not reported Not reported	TC1-3/IC1-3: HR, 0.71; 95% CI, 0.55–0.91 TC3/IC3[a]: HR, 0.76; 95% CI, 0.49–1.17 TC ≥1%[a]: HR, 0.66; 95% CI, 0.50–0.87 TC ≥50%: HR, 0.59; 95% CI, 0.39–0.90
IMpower 131, Jotte et al,[174] 2020	Y	Atezolizumab + chemotherapy, n = 343 Chemotherapy, n = 340	SP142	Not reported	TC2/3 or IC2/3: HR, 0.72; 95% CI, 0.52–1.00 TC3 or IC3[a]: HR, 0.48; 95% CI, 0.29–0.81
IMpower 132, Nishio et al,[175] 2021	Y	Atezolizumab + chemotherapy, n = 292 Chemotherapy, n = 286	SP142	Not reported	TC1/2 or IC1/2: HR, 1.18; 95% CI, 0.80–1.76 TC3 or IC3[a]: HR, 0.73; 95% CI, 0.31–1.73

Table 3. Continued

RCT	FDA Approval, Y/N	Treatment Arms	PD-L1 IHC Clone	Overall Response Rate	Overall Survival
IMpower 130, West et al,[20] 2019	Y	Atezolizumab + chemotherapy, n = 483 Chemotherapy, n = 240	SP142	Not reported	TC1/2 or IC1/2: HR, 0.70; 95% CI, 0.45–1.08 TC3/IC3: HR, 0.84; 95% CI, 0.51–1.39

Abbreviations: CheckMate, an open-label, randomized phase 3 trial of nivolumab, or nivolumab plus ipilimumab, or nivolumab plus platinum doublet chemotherapy versus platinum doublet chemotherapy in subjects with chemotherapy-naïve stage IV or recurrent non–small cell lung cancer; FDA, US Food and Drug Administration; HR, hazard ratio; IC, immune cell; ICS, immune cell score; IHC, immunohistochemistry; IMpower, a phase III, open label, randomized study of atezolizumab (anti–PD-L1 antibody) compared with a platinum agent (cisplatin or carboplatin) in combination with either pemetrexed or gemcitabine for PD-L1–selected, chemotherapy-naive patients with stage IV nonsquamous or squamous non–small cell lung cancer; KEYNOTE, study of pembrolizumab (MK-3475) versus platinum-based chemotherapy for participants with PD-L1–positive advanced or metastatic non–small cell lung cancer; N, no; OR, odds ratio; PD-L1, programmed death ligand-1; RCT, randomized control trial; TC, tumor cell; TPS, tumor proportion score; Y, yes.

[a] FDA-approved TPS/ICS cutoff.

contrast to low response rates for immunotherapy in patients with *EGFR* and *ALK* alterations, patients with tumors harboring *BRAF* mutations appear to respond just as well to immunotherapy as those with wild-type tumors, whereas data on outcomes of immunotherapy in patients with other targetable driver alterations (eg, *ROS1*, *RET*, *MET*) are relatively limited and inconclusive.[50–52] Patients with targetable oncogene-driven NSCLC derive substantial benefit from appropriate targeted therapy in the first-line setting,[53–55] arguing for the value of obtaining comprehensive genomic profiling at the time of diagnosis with advanced disease.[56]

During the public comment period, of a total of 79 responders, 73 (94.80%) agreed or agreed with suggested modifications to the draft statement, 4 (5.19%) disagreed, and 2 (2.53%) were neutral. There were 17 written comments, many of which suggested that the final recommendation statement does not include the word *advanced*. The general comments also suggested that the FDA approval was for early stage, although the evidence included in the data review was on patients with late-stage NSCLC. Public comments also suggested that a discussion of the utility of PD-L1 in addition to a complete lung molecular biomarker profile is important. These comments were taken into consideration. While the recommendation remained the same, the comments were addressed in the discussion above.

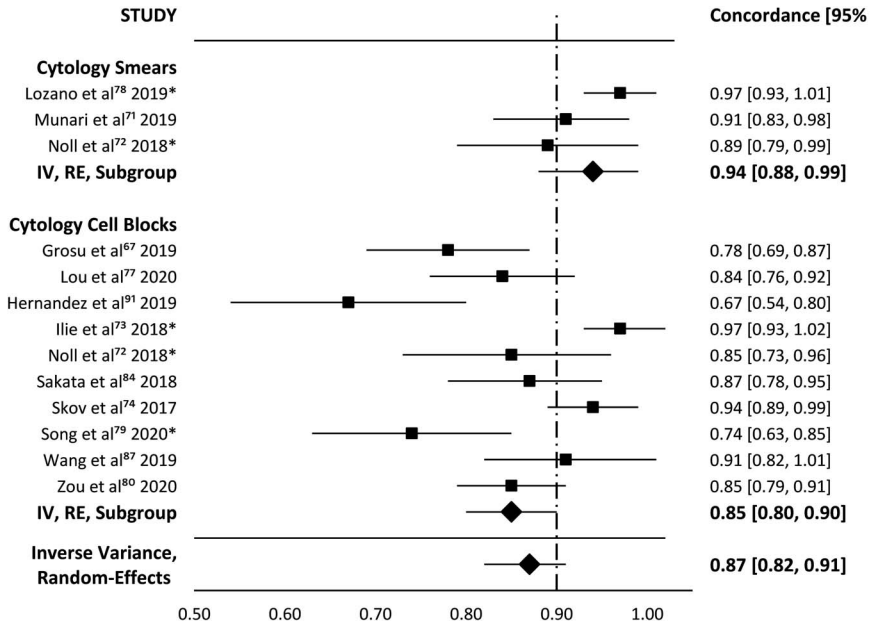
2. Pathologists should ensure appropriate validation has been performed on all specimen types and fixatives.

Note: Specific validation requirements are out of scope with this guideline, and laboratories should refer to the *Principles of Analytic Validation of Immunohistochemical Assays Guideline*[57] for details on how to validate IHC specimens.

(Strength of Recommendation: Conditional; Certainty of Evidence: Low)

The evidence base informing this statement is composed of 35 studies reporting on immunotherapy RR and survival rates,[22,58] PD-L1 status concordance (Figure),[59–85] diagnostic test characteristics of PD-L1 expression detection,[71,77,82–84] and PD-L1 status using various specimen types.[86,87] Both interobserver and intraobserver agreement for PD-L1 status using tissue[69,75,88–90] and cytology specimens[69,71,75,76,91] were also considered. The certainty of evidence across the 19 outcomes ranged from very low through moderate. EP members defined the benefits of PD-L1 expression detection using the best specimen available as moderate; however, the harms were also defined as moderate and the overall certainty of evidence was low, leading to the conclusion that balance of effects probably favored testing in the best available specimen. EP members also discussed that there was possibly important variability in the values and preferences of key stakeholders, but the guidance is expected to be acceptable and feasible to implement. Refer to Supplemental Tables 5 through 8 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement. Supplemental Table 3 summarizes the EtD framework.

Several variables may affect a given specimen's suitability for testing, including age, site of origin, size (biopsy versus resection), and modality (histologic versus cytologic preparation). While several studies examining PD-L1 score concordance in relation to these variables were reviewed, only a handful analyzed clinical outcomes and response to ICI therapy. Based on the available evidence, it is difficult to



PD-L1 tumor proportion score in cytology specimens versus histology sections. Reference standard defined as surgical resection for all studies except those denoted with an asterisk. In asterisk-denoted studies, the reference standard was a mixed FFPE sample of cell blocks, small biopsy samples, and surgical sections. Abbreviations: FFPE, formalin-fixed, paraffin-embedded; IV, inverse variance; PD-L1, programmed death ligand-1; RE, random effects.

determine what constitutes an optimal specimen for testing in patients with advanced NSCLC. Given that acquisition of material for testing entails a degree of morbidity depending on the patient's condition and distribution of disease, the recommendation provides room for clinical judgment on the part of the pathologist and treating oncologist about selection of the most appropriate specimen, particularly when multiple specimens are available for testing.

The minimum criteria for adequacy as defined by the assay description for the PD-L1 IHC 22C3 Dako/Agilent pharmDx assay (Agilent Technologies, Inc, Santa Clara, California) is 100 viable cells.[92] This minimum threshold is supported by 2 studies that demonstrate higher rates of positivity at both the 1% and 50% TPS cutoffs[93,94] in cases where at least 100 viable cells are available for review. In specimens that fail to meet this threshold, deeper levels from the paraffin tissue block may yield adequate cellularity or an alternative specimen may be evaluated if available. If the initial specimen is negative, consideration should be given to repeating the test on another specimen if available.

Testing of the primary tumor versus metastatic sites was also considered by the EP. The available evidence in the literature demonstrates that the TPS scores obtained from the primary tumor and synchronous metastatic lesions are discordant at both the 1% and 50% cutoffs in resected specimens approximately 20% to 30% of the time.[59,61,64,65] There were no studies evaluating the RR to PD-1 or PD-L1 ICIs, based on testing specimens from different sites available for review; therefore, the EP could not make a recommendation regarding the testing of a primary tumor versus a synchronous metastatic lesion. When synchronous specimens from multiple sites are available, a reasonable approach would be to test the specimen of the best technical quality (ie, highest viable cellularity and best preservation).

Another important consideration in specimen selection for PD-L1 IHC testing is the potential for discordant results when testing small biopsy samples versus resection specimens. Intratumoral heterogeneity of PD-L1 expression is frequently seen in resection specimens; therefore, it is possible that results obtained from small biopsy samples may differ depending on what region of a tumor is sampled. Retrospective evidence[95–97] suggests there is potential variation in the degree of PD-L1 expression in different tumoral patterns, in both lung adenocarcinoma and lung squamous cell carcinoma.[95] As a good practice, the EP suggests interpreting PD-L1 TPS with caution when performed on a primary lung biopsy specimen that may contain limited representation of the overall tumor growth, such as a biopsy specimen containing only lepidic component. Given that patients with advanced NSCLC who are candidates for ICI therapy will in most cases not undergo resection of the primary tumor or a metastatic lesion, the performance characteristics of PD-L1 IHC using more limited samples (eg, core biopsies, cytology specimens, and endobronchial biopsies) versus resected specimens can only be extrapolated from retrospective analyses of predominantly early-stage tumors. The evidence demonstrates that concordance between small biopsy samples and resected specimens ranges from 83% to 95% at the 50% cutoff and 77% to 86% at the 1% cutoff.[82–84] In most discordant cases, a lower score is obtained from the small biopsy sample than from the corresponding resection specimen.[98] Based on these findings, testing of small biopsy samples is acceptable.

Many patients with NSCLC, particularly those with advanced disease, are diagnosed by fine-needle aspiration or other cytologic techniques, and such specimens may be the only available pathology material. Studies comparing PD-L1 expression on tumor cells in cytology cell blocks from various sources—including both needle aspirate specimens and body fluids—to expression in paired resection specimens[67,70,74,75,77,79,80] show good concordance, ranging from 67% to 94% with a pooled concordance of 85% (95% CI, 0.80–0.90)[99] (Figure). A small number of studies have evaluated the use of PD-L1 immunocytochemistry performed on alcohol-fixed cytology material. Noll et al[72] and Lozano et al[78] compared TPS obtained from alcohol-fixed aspirate slides to that obtained from concurrent histologic core needle biopsies and found an overall concordance rate of 97% (Lozano) and 89% (Noll) at the clinically relevant cutoffs of 1% and 50% (Figure). Of note, Noll et al[72] used the 22C3 Dako/Agilent

pharmDx clone, while Lozano et al[78] used both the VENTANA PD-L1 (SP263) assay (Roche Diagnostics, Indianapolis, Indiana) and the 22C3 clone in their series of cases. Munari et al[71] compared PD-L1 scores obtained from ex vivo specimen aspirates and paired whole sections from resections, using the VENTANA BenchMark ULTRASP263 clone, and reported a concordance rate of 81% at the 1% cutoff and 91% at the 50% cutoff (Figure). In most discordant cases, the score obtained from the aspirate smear was lower than that from the paired resection specimen. Of note, both the SP263 and 22C3 assays designate that they are clinically validated for use in formalin-fixed, paraffin-embedded (FFPE) material. While the limited data currently available for review appear promising, at the current time, alcohol-fixed cytologic material should be used for PD-L1 testing only if FFPE materials are not available, and in laboratories that have specifically validated this sample type, and the results should be reported with a comment that PD-L1 testing in non-FFPE tissues has not been clinically validated.

Previously collected archival tissue is often the most readily available source for biomarker testing and mitigates the need for rebiopsy in patients who may not be amenable to further intervention. However, some studies have raised issues regarding antigen decay in archival tissue, particularly nuclear and membranous staining,[57,100,101] which may have implications for PD-L1 TPS. In fact, preliminary studies suggest that the PD-L1 clone 22C3 is prone to deglycosylation and diminished expression after a year or more in storage.[102] Additionally, tumor expression of PD-L1 is dynamic, unlike other predictive biomarkers, with some discordance in expression reported following treatment between initial and recurrent disease.[103] In fact, the study of 2 doses of pembrolizumab (MK-3475) versus docetaxel in previously treated participants with non–small cell lung cancer, or KEYNOTE-010,[104] was amended to require PD-L1 evaluation in newly collected samples except when patient safety posed a significant risk. As such, formal evaluation of the utility of archival tissue in this context is of practical importance. At this time, data are limited to a study by Herbst et al,[22] which compared PD-L1 status and response rate in archived samples and fresh biopsy samples, using the 22C3 pharmDx assay. In this study, 455 archival samples were evaluated with a median time between sample collection and PD-L1 testing of 250 days (range, 3–2510 days). Five hundred seventy-eight "new" samples were evaluated that had an average of 11 days between sample collection and PD-L1 testing (range, 1–371 days). The results of this study showed that PD-L1 expression levels with TPS of 1% or more and TPS of 50% or more were similar between archival and newly collected samples. The authors[22] concluded that PD-L1 expression was adequately preserved following a median 8 months of storage and that expression was potentially not affected by intervening treatment. Additionally, OS and progression-free survival hazard ratios (HRs) were similar across archival and newly collected samples in both 1%-or-more and 50%-or-more TPS cohorts.

Bone is a frequent site of metastasis for lung cancer; hence, the impact of decalcification methods on PD-L1 expression is of key interest. Three studies[50,105,106] evaluated the expression of the 22C3 Dako/Agilent pharmDx assay in decalcified tissue samples; Forest et al[50] also included the E1L3N (PD-L1 [E1L3N] XP, Cell Signaling Technology, Danvers, Massachusetts) clone. Strickland et al[105] evaluated PD-L1 expression following decalcification with EDTA, formic acid/MasterCal IM Plus (FA/MC), 12% hydrochloric acid (HCl), and Decal STAT Decalcifier/23% HCl solutions at periods of 1, 2, 6, and 24 hours. This study found that EDTA and FA/MC had little effect on PD-L1 expression, whereas 12% HCl resulted in a progressive decline in expression. Notably, Decal STAT dramatically reduced expression at all treatment durations. Forest et al[50] evaluated PD-L1 expression following EDTA or acid decalcification and did not detect a statistically significant difference in reactivity with the E1L3N clone for the first 24 hours or after delayed fixation following EDTA decalcification. Labeling of carcinoma cells by the 22C3 pharmDx assay was slightly decreased following EDTA decalcification (an observation that did not reach statistical significance); in contrast, there was a significant decrease in percentage of tumor cell staining following acid decalcification. Pontarollo et al[106] evaluated PD-L1 expression following EDTA or formic acid decalcification and did not detect a significant difference in TPS in decalcified versus nondecalcified specimens. Available data suggest that specimens decalcified in EDTA or formic acid for less than 24 hours may generate PD-L1 TPS scores that are comparable to those of specimens exposed to formalin alone; however, nondecalcified samples should be prioritized for PD-L1 testing, when available, and laboratories should specifically validate decalcified specimens if they offer testing for this sample type. The use of decalcified specimens for PD-L1 testing to select patients for ICI therapy has not been clinically validated.

The public comment period recorded 78 responders, of whom 66 (84.62%) agreed or agreed with suggested statement modifications, 6 (7.69%) disagreed, and 6 (7.69%) were neutral. There were 10 written comments, many of which suggested that the final recommendation language be changed from "clinician" to "pathologist." Suggested comments also included a request to clarify the definition of "best available specimen." All comments were taken into consideration. The recommendation statement was edited to reflect that the pathologist is the intended subject of the recommendation. In addition, the statement was modified to remove language around the best available specimen and instead emphasizes that validation needs to occur for all potential specimen types and fixatives. The discussion text includes an explanation of the variables that make defining an optimal specimen for testing difficult, and the discussion text includes an explanation of the best available specimen, as well as a recommendation to use available guidelines for appropriate validation.

3. When feasible, pathologists should use clinically validated PD-L1 immunohistochemistry assays as intended. (Strength of Recommendation: Conditional; Certainty of Evidence: Very Low)

The evidence base supporting this statement includes 54 studies reporting on PD-L1 status concordance,[37,38,74,78,88,107–120] diagnostic test characteristics,[108] and interobserver agreement[112,116,121–126] of various combinations of clinically validated PD-L1 assays. Additionally, clinical trials and observational studies reporting on immunotherapy survival and RR,[10–38,104,119,127] leading to the clinical validation of the assays, were used as indirect evidence to support this statement. Certainty of evidence for the outcomes was assessed as low and very low. The EP members discussed clinically validated, FDA-approved PD-L1 IHC assays (CDx assays for the purpose of this document) versus laboratory-developed and validated PD-L1 IHC assays at length. From the available evidence, the EP members determined that use of CDx assays carried moderate benefits; however, the harms of CDx assays, including availability of the testing platforms and clones and specific training for each CDx, were also defined as moderate. The EP members concluded that the balance of effects did not favor either CDx assays or

Table 4. FDA Approval Criteria for PD-L1 Companion Diagnostic Assays			
IHC Clone	Platform	PD-L1 Protein Expression Cut Point	Anti–PD-1/PD-L1 Agent
22C3	Dako/Agilent	TPS >1%	Pembrolizumab monotherapy
		TPS ≥50%	Cemiplimab
28-8	Dako/Agilent	TPS ≥1%	Nivolumab
			Nivolumab plus ipilimumab
SP142	VENTANA	TPS ≥50% or ICS ≥10%	Atezolizumab
SP263	VENTANA	TCs ≥1%	Atezolizumab

Abbreviations: FDA, US Food and Drug Administration; ICS, immune cell score; IHC, immunohistochemistry; PD-1, programmed death receptor-1; PD-L1, programmed death ligand-1; TCs, tumor cells; TPS, tumor proportion score.

laboratory-developed tests (LDTs). The EP members also determined that use of CDx assays carried a large cost and could lead to reduced health equity. A *conditional* strength of recommendation was based on the clinical validation of these assays and their established ability to predict immunotherapy response but with an understanding of the limitations of this guidance. EP members agreed that the guidance would probably be acceptable to key stakeholders and probably feasible to implement. Refer to Supplemental Tables 5 through 8 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement, Supplemental Table 3 summarizes the EtD framework.

For PD-L1 testing, there are 4 CDx assays—PD-L1 IHC 22C3 Dako/Agilent pharmDx, PD-L1 IHC 28-8 Dako/Agilent pharmDx, VENTANA PD-L1 SP142, and SP263 assays—each of which was codeveloped with a specific anti–PD-1/PD-L1 agent used in clinical trials and has been approved by the FDA and/or other regulatory agencies as companion diagnostics for the use of those agents (for intended use) in NSCLC. Each CDx assay is determined by a specific combination of an antibody clone, IHC testing platform, and reagents (Table 4). Thus, any PD-L1 IHC assay that consists of different combinations from those of the 4 FDA-approved assays, or (as noted earlier) is used on tissues fixed in a manner not included in the complementary clinical trial(s), is considered an LDT (*see Recommendation 4*). For financial and/or practical reasons, it may be difficult for a given pathology laboratory to be equipped with multiple CDx assays for PD-L1 testing, and the laboratory may opt to use 1 or few assays, either CDx or LDTs, to determine the eligibility for treatment with various anti–PD-1/PD-L1 agents.[128,129]

In this context, it is important to note that the analytic compatibility and exchangeability of PD-L1 IHC assays for tumor cell testing, in particular those of the 4 CDx assays, have been extensively studied.[37,38,74,78,88,107–120] Most studies reported similar results—expression concordance between 22C3, 28-8, and SP263 CDx assays is typically 90% or greater, while the VENTANA PD-L1 SP142 CDx generally shows weaker expression than the other 3 CDx assays with fewer positive tumor cells[37,115] (Tables 5 and 6). As for reproducibility, interobserver agreements around tumor cell scoring of PD-L1 are similar among the 4 CDx assays and generally substantial to almost perfect for both 1% and 50% cutoffs,[112,116,121–126] whereas those on immune cells are only slight or fair.[125,126] The technical performance and interobserver agreements of LDTs will be discussed in Recommendation 4.

Fitzgibbons et al[57] state that, for initial validation of every assay used clinically, laboratories should achieve at least 90% overall concordance between the new assay and the comparator assay or expected results. Given its failure to meet the guideline recommendations, the SP142 CDx is not considered appropriate for the selection of patients for treatment with pembrolizumab, cemiplimab, or nivolumab/ipilimumab. While data suggest that 22C3, 28-8, and SP263 CDx assays may be interchangeable, information about predictive performance of those CDx assays for anti–PD-1/PD-L1 agents outside of their given indication or that of LDTs is limited.[37] The same is true of LDTs, based on the primary antibodies in these CDx assays and on other anti–PD-1/PD-L1 clones. If feasible, pathologists should use CDx assays as intended, until larger cohort studies confirm the comparability of those CDx assays from both technical and clinical standpoints. The FDA has embraced more generic language in certain therapeutic approvals,[130] indicating that PD-L1 status "as determined by an FDA-approved test" may be used for patient selection. The laboratory director should be familiar with these testing indications and approvals when determining which assays to validate for clinical use. The CDx assays if used outside of their trial-defined indications require that they undergo a technical validation on par with any LDT as defined by the CAP validation guidelines.

Table 5. PD-L1 Status Concordance in FDA-Approved Companion Diagnostic Assays and Laboratory-Developed Tests (LDTs)				
FDA-Approved CDx[a]	LDT (Clone; Platform)	TPS Cutoff		Concordance (Range Reported by Included Studies)
		CDx	LDT	
22C3 pharmDx	73-10; Dako/Agilent	≥50%	≥80%	93.9%, Grote et al,[131] 2020
	E1L3N; VENTANA	≥50%	≥50%	88.2%, Munari et al,[124] 2019
SP263 assay	22C3; VENTANA	≥1%	≥1%	89.7%–100%, Munari et al,[132] 2018, Sughayer et al,[136] 2019
	E1L3N; VENTANA	≥50%	≥50%	95.8%, Munari et al,[124] 2019

Abbreviations: CDx, companion diagnostic assay; FDA, US Food and Drug Administration; PD-L1, programmed death ligand-1; TPS, tumor proportion score.

[a] FDA approval criteria are outlined in Table 4.

Table 6. Reported Kappas for PD-L1 Status in FDA-Approved Companion Diagnostic Assays and Laboratory-Developed Tests (LDTs)				
FDA-Approved CDx[a]	LDT (Clone; Platform)	TPS Cutoff		κ (Range Reported by Included Studies)
		CDx	LDT	
SP263 assay	22C3; VENTANA	≥50%	≥50%	0.73–0.75, Munari et al,[132] 2018
	SP263; Dako/Agilent	≥1%	≥1%	0.83–0.86, Adam et al,[134] 2018
	SP263; Leica	≥1%	≥1%	0.83–0.86, Adam et al,[134] 2018
	SP142; Dako/Agilent	≥1%	≥1%	0.38–0.68, Adam et al,[134] 2018
	SP142; Leica	≥1%	≥1%	0.78–0.81, Adam et al,[134] 2018
	E1L3N; Dako/Agilent	≥1%	≥1%	0.63–0.77, Adam et al,[134] 2018
	E1L3N; VENTANA	≥1%	≥1%	0.60–0.81, Adam et al,[134] 2018
	E1L3N; Leica	≥1%	≥1%	0.75–0.78, Adam et al,[134] 2018
SP142 assay	E1L3N; VENTANA	≥1%	≥1%	0.65, Kim et al,[135] 2017
22C3 assay	22C3; VENTANA	≥1%	≥1%	0.77–0.81, Adam et al,[134] 2018
	22C3; Leica	≥1%	≥1%	0.50–0.62, Adam et al,[134] 2018
28-8 assay	28-8; VENTANA	≥1%	≥1%	0.73–0.80, Adam et al,[134] 2018
	28-8; Leica	≥1%	≥1%	0.58–0.60, Adam et al,[134] 2018

Abbreviations: CDx, companion diagnostic assay; FDA, US Food and Drug Administration; PD-L1, programmed death ligand-1; TPS, tumor proportion score.

[a] FDA approval criteria are outlined in Table 4.

During the public comment period, of a total of 79 responses, 70 (88.61%) agreed or agreed with suggested modifications to the statement, 4 (5.06%) disagreed, and 5 (6.33%) were neutral. There were 28 written comments. Some comments suggested using the term *pathologist* instead of *laboratories* for clarity. In addition, there were several comments about the use of the text "clinically validated" in the statement. While the final statement did not reflect this suggested change, this is discussed within many sections addressing validation. The public commented on the importance of clinically validated assays and their utility as intended by the assay manufacturer. Although validation is discussed in the text, the details on how to validate assays and general quality assurance measures are out of scope for this guideline. All comments were taken into consideration. The final recommendation statement was edited to clarify that it is the pathologist carrying out the recommendation.

4. Pathologists who choose to use laboratory-developed tests for PD-L1 expression should validate according to the requirements of their accrediting body.

(Strength of Recommendation: Strong; Certainty of Evidence: Very Low)

The evidence base informing this statement is composed of 1 study reporting on immunotherapy RRs using clones 22C3 and 73-10,[131] and 8 studies reporting on PD-L1 status concordance,[73,124,131–136] diagnostic test characteristics,[124] and interobserver agreement[124,125] of LDTs when compared with clinically validated IHC assays. The certainty of evidence was low and very low for the outcomes of interest. From the available evidence, however, EP members defined the benefits of validating all LDTs as large and the harms of the validation as trivial and thus concluded that the benefits outweighed the harms. It is expected that this guidance will be acceptable to key stakeholders and feasible to implement. In addition, given the possible variable performance of unvalidated LDTs when compared to CDx-based PD-L1, the EP members concluded that not performing required validation[137] on LDTs could lead to substantial harms to patients, leading to this recommendation as *strong*, despite a certainty of evidence rating of very low. Refer to Supplemental Tables 5 through 8 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement. Supplemental Table 3 summarizes the EtD framework.

An LDT is defined as an in vitro diagnostic test that is designed and used within a single laboratory.[138,139] The FDA notes that LDTs are important to the continued development of personalized medicine, and that in vitro diagnostics should be accurate so that patients and health care providers do not seek unnecessary treatments, delay needed treatments, or become exposed to inappropriate therapies. An LDT may use one of the available anti–PD-L1 clones used for CDx or another clone (such as E1L3N) that has never been validated in prospective clinical trials, in combination with any available IHC platform[128] (Tables 7 and 8). Laboratories may run LDTs for financial or practical reasons, but as the FDA noted, it is important that they be accurate. Thus, all LDTs should be validated. LDTs including those with clone E1L3N can reasonably match the technical performance of CDx assays,[73,124,131–133,135,136,140] but multi-institutional studies evaluating a large number of LDTs have reported that only 50% to 60% were analytically compatible to CDx assays.[134,141] As for reproducibility, interobserver agreement on tumor cell scoring of PD-L1, using clone E1L3N LDTs among multiple pathologists, was similar to that using 22C3, SP263, and SP142 CDx assays for both 1% and 50% cutoffs.[124,125] While specific validation requirements are out of scope with this guideline, technical validation of an LDT using a CDx as the gold standard[142] is important for the appropriate selection of patients for treatment with PD-1 axis blockade.

The PD-L1 IHC 73-10 Dako/Agilent pharmDx assay, a combination of the clone 73-10 and a Dako/Agilent platform, has not yet been approved by the FDA, but it was codeveloped with an anti–PD-L1 agent, avelumab, and has been evaluated in multiple clinical trials of patients with NSCLC.[143,144] Therefore, the 73-10 IHC assay is distinct from other LDTs, since it is used globally beyond a single laboratory. Data on the analytic performance of the 73-10 IHC assay are scarce, likely owing to its limited accessibility.[128] Reportedly, the 73-10 assay is more sensitive than the 4 CDx assays.[126] The 73-10 pharmDx assay

Table 7. Laboratory-Developed Tests Using FDA-Approved Antibodies Described in the Literature		
Approved[a] Assay	Modified PD-L1 Expression Cut Point	Modified Platform
22C3 assay	TPS 1%–49%, Gadgeel et al,[170] 2020 Herbst et al,[41] 2021	VENTANA, Munari et al,[132] 2018, Villaruz et al,[133] 2019, Sughayer et al,[136] 2019, Ilie et al,[73] 2018, Adam et al,[134] 2018
	TPS ≥50%, Reck et al,[39] 2021, Mok et al,[17] 2019, Gadgeel et al,[170] 2020, Herbst et al,[41] 2021	Leica, Adam et al,[134] 2018
28-8 assay	TPS ≥5%, Carbone et al,[172] 2017	VENTANA, Adam et al,[134] 2018
	TPS ≥50%, Brahmer et al,[13] 2015, Hellmann et al,[16] 2019	Leica, Adam et al,[134] 2018
SP142 assay	TC1/2 or IC1/2, West et al,[20] 2019	Dako/Agilent, Adam et al,[134] 2018
	TC2/3 or IC2/3, Herbst et al,[119] 2020	Leica, Adam et al,[134] 2018
	TC1-3 or IC1-3, Socinski et al,[173] 2021	
SP263 assay	None reported	Dako/Agilent, Adam et al,[134] 2018
		Leica, Adam et al,[134] 2018

Abbreviation: FDA, US Food and Drug Administration; IC, immune cells; PD-L1, programmed death ligand-1; TC, tumor cells; TPS, tumor proportion score.

[a] FDA approval criteria are outlined in Table 4.

was optimized to identify tumors with low PD-L1 expression, susceptible to antibody-dependent cellular cytotoxicity through a wild-type Fc region—a unique property of avelumab along with PD-1 axis blockade.[143] A study with 231 NSCLC tissue samples, including 83 from a clinical trial for avelumab, showed overall similar analytic and predictive performances along with a higher negative predictive value of the 73-10 assay as compared to those of the 22C3 CDx assay when the cutoffs of 1%, 50%, and 80%, and the cutoffs of 1%, 20%, and 50%, respectively, were compared.[131,145] Given the differences in analytic cutoffs used to develop 73-10, compared to those for the other CDx assays, additional studies are warranted to evaluate the analytic compatibility of these assays more carefully.

During the public comment period, of a total of 76 responders, 66 (86.84%) agreed or agreed with suggested modifications to the draft statement, 3 (3.95%) disagreed, and 7 (9.21%) were neutral. There were 10 written comments; some agreed that adequate validation is a must when using an LDT assay, while there were some who advocated for the use of only FDA-approved assays. In addition, there were comments about the low certainty of evidence with a strong recommendation. The final recommendation was modified to reinforce that LDTs are appropriate to use if the assays are adequately validated according to the laboratory's accreditation requirements. To address the strong recommendation for low levels of certainty of evidence, the EtD framework used by the EP further reinforced the importance of validation with the use of LDT assays.

5. Pathologists should report PD-L1 immunohistochemistry results using a percentage expression score.

(Strength of Recommendation: Conditional; Certainty of Evidence: Very Low)

The guideline statement is supported by 5 studies reporting on immunotherapy RRs[17,22,29,31,127] and survival rates[17,22,29,31,127] stratified by specific PD-L1 TPS thresholds. An additional 5 studies evaluated interobserver agreement, using multiple IHC clones also stratified by TPS score.[112,116,122–124] The certainty of evidence for RRs was assessed as low, based on a very serious aggregate risk of bias across the studies reporting on the outcome, while the certainty for survival and interobserver agreement was assessed as very low owing to very serious risk of bias plus downgrading for inconsistency (Supplemental Table 9). From the available evidence, EP members concluded that reporting PD-L1 expression as a percentage score carried moderate benefits and only small harms, leading to the determination that benefits probably outweighed the potential harms. This recommendation is expected to be acceptable to key stakeholders and will be feasible to implement. This guidance is expected to have no impact on health equity, and the resource requirements were considered to be negligible. Refer to Supplemental Tables 5 through 9 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement. Supplemental Table 3 summarizes the EtD framework. The levels of PD-L1 TPS demonstrate statistically significant correlation with patient response and survival following

Table 8. Laboratory-Developed Tests Using Antibodies Not FDA Approved			
IHC Clone	Platform	PD-L1 Protein Expression Cut Point	Anti–PD-1/PD-L1 Agent
73-10	Dako/Agilent	TPS ≥1%, Barlesi et al,[127] 2018, Grote et al,[131] 2020, Park et al,[40] 2021	Avelumab, Barlesi et al,[127] 2018, Grote et al,[131] 2020, Park et al,[40] 2021
		TPS ≥50%, Barlesi et al,[127] 2018, Grote et al,[131] 2020, Park et al,[40] 2021	
		TPS ≥80%, Barlesi et al,[127] 2018, Grote et al,[131] 2020, Park et al,[40] 2021	
E1L3N	VENTANA	TPS ≥1%, Kim et al,[135] 2017, Munari et al,[124] 2019	Atezolizumab, Kim et al,[135] 2017, Munari et al,[124] 2019
		TPS ≥5%, Kim et al,[135] 2017	
		TPS ≥50%, Kim et al,[135] 2017, Munari et al,[124] 2019	

Abbreviations: FDA, US Food and Drug Administration; IHC, immunohistochemistry; PD-1, programmed death receptor-1; PD-L1, programmed death ligand-1; TPS, tumor proportion score.

immunotherapy with ICI administered in isolation and in chemo-immunotherapy combinations. Published evidence demonstrates higher RR and improved outcomes in patients with higher levels of PD-L1 expression, particularly among those patients with high PD-L1 TPS (at least 50%), both in the context of retrospective correlative studies using "real-life data" and in the context of RCTs.[21,46,90,104,146–148] For this reason, the EP recommends that pathologists report PD-L1 IHC results by using a percentage expression score.

From a practical standpoint, providing an exact percentage expression score value may be challenging owing to the subjective nature of visual assessment of PD-L1 expression and scoring variability among pathologists, particularly regarding assessment of immune cell populations. Recent publications have shown high interpathologist concordance when scoring tumor cells, but lower agreement on immune cell scoring across different PD-L1 assays.[90,125,127] In addition, other PD-L1 test–related variables such as sample selection and availability, differences between PD-L1 assays, and intratumor heterogeneity of PD-L1 expression may influence the ability of pathologists to consistently provide precise quantitative measurements. For this reason, one option that several clinical laboratories have adopted is to report ranges of PD-L1 percentage expression scores (eg, 5% or 10% incremental values) instead of absolute scores. This semiquantitative approach is expected to be more accurate and reproducible than reporting specific expression percentage values and still provides sufficient information for management decisions, provided that ranges are reported with management-based cut points in mind. A more objective quantification of PD-L1 expression could conceivably be achieved with the use of assay-specific controls representing expression intensities across the dynamic range of the test.[105]

A total of 77 responses were received during the public comment period, of which 65 (84.42%) agreed or agreed with suggested modifications to the draft statement, 3 (3.90%) disagreed, and 9 (11.69%) were neutral. There were 27 written comments. The most common comments to the draft statement included using percentage cutoffs by assay, and how scoring is dependent on specific clone. The final recommendation was slightly edited without change to the context. All comments received were reviewed and addressed with the discussion.

6. Clinicians should not use tumor mutation burden alone to select patients with advanced NSCLC for immune checkpoint inhibitors, based on insufficient evidence in this population.

(Strength of Recommendation: Conditional; Certainty of Evidence: Very Low)

The evidence base is composed of 8 studies reporting on immunotherapy RRs[149,150] and survival rates[29,32,35,149–153] when correlated with TMB status. Certainty of evidence was assessed as very low for RRs and very low for survival rates. This conditional recommendation was based on the trivial benefits of using TMB to select patients for immunotherapy paired with the moderate harms of its use. The balance of effects favored not using TMB, as did the moderate costs and probable reduced health equity that would be associated with recommending its use. Further to these domains, the EP members concluded that guidance in support of TMB would not be acceptable to key stakeholders and probably not be feasible to implement. Refer to Supplemental Tables 5 through 8 for a summary of the risk of bias assessment for all included studies and the certainty of evidence assessment for all outcomes informing the statement. Supplemental Table 3 summarizes the EtD framework.

Recommendation statement 6 regarding the use of TMB received a conditional strength based on low and very low certainty of evidence for impact on survival and RRs, respectively. Regarding the line of therapy that is relevant to this recommendation, the 8 studies evaluated in this evidence base include heterogeneous lines of administration of therapy, including in the context of first-line therapy[152,153]; a combination of lines from first through third[35,149]; palliative setting[29]; and undisclosed treatment setting.[32,151]

Despite initial findings from the CheckMate-227[16] trial showing an improved 1-year progression-free survival rate for patients with advanced NSCLC and TMB of 10 mutations/megabase (mut/Mb) or greater and receiving nivolumab plus ipilimumab in the first-line setting,[154] the supplemental FDA application was withdrawn when subsequent data showed no difference in survival outcomes between patients stratified by high or low tumor TMB. The median OS with nivolumab + ipilimumab in patients with TMB of 10 mut/Mb or greater was 23.03 months versus 16.72 months for the chemotherapy arm (HR, 0.77; 95% CI, 0.56–1.06); among patients with TMB lower than 10 mut/Mb, the median OS was 16.20 months versus 12.42 months, respectively (HR, 0.78; 95% CI, 0.61–1.00). At present, the FDA approval for nivolumab plus ipilimumab as first-line treatment for patients with metastatic NSCLC requires tumor PD-L1 expression of 1% or more, as determined by an FDA-approved test, with no *EGFR* and *ALK* genomic tumor aberrations, and does not include TMB.

Accelerated approval was granted for pembrolizumab in the treatment of adult and pediatric patients with unresectable or metastatic TMB-high (≥10 mut/Mb) solid tumors—as determined by an FDA-approved test—that have progressed following prior treatment and who have no satisfactory alternative treatment options. Although this is a tumor-agnostic approval, it is worth noting that the KEYNOTE-158 study[155] (the basis of this approval) included 102 patients with TMB of 10 mut/Mb or greater spanning 9 different tumor types, none of which were NSCLC.[156] These data came from an analysis of 10 cohorts of patients with various previously treated, unresectable or metastatic solid tumors, and response rates (not survival rates) were compared against those who did not have high TMB.

A retrospective study[157] evaluating 909 nonsquamous NSCLCs, with both PD-L1 and TMB data available, identified that the median TMB was significantly higher in the PD-L1–high group than in the PD-L1–low and negative groups (median, 12.2 mut/Mb versus 10.6 mut/Mb versus 10.6 mut/Mb, respectively; $P < .001$) with a modest, but significant linear correlation between TMB and PD-L1 TPS ($P = .12$; $P < .001$). Additionally, in a multivariable logistic regression analysis evaluating factors associated with response to ICIs, improved progression-free survival was significantly associated with both higher PD-L1 expression (HR, 0.41 [95% CI, 0.28–0.62]; $P < .001$) and TMB (HR, 0.97 [95% CI, 0.95–0.99]; $P = .002$), whereas OS was significantly associated only with PD-L1 expression (HR, 0.59 [95% CI, 0.38–0.93]; $P = .02$).

The open comment period collected a total of 78 responders, of whom 59 (75.64%) agreed or agreed with suggested modifications to the draft statement, 4 (5.13%) disagreed, and 15 (19.23%) were neutral. There were 12 written comments. These included the recent FDA approval of pembrolizumab with the use of TMB-high. Other comments included the use of TMB-high only if treatment options are not available. The EP reviewed all comments and decided that no edits are necessary, although the FDA approval should be part of the discussion text.

LIMITATIONS

PD-L1 expression and TMB testing are the most widely used biomarkers in patients with NSCLC being considered for ICI therapy. Despite widespread adoption, findings from the extensive literature review informing this guideline demonstrate that conclusions about several PD-L1 and TMB test-related variables are difficult to draw owing to the limited number of publications addressing specific technical aspects associated with these biomarkers. Notwithstanding these limitations and the complexity surrounding testing of these biomarkers, the EP developed evidence-based recommendations that address preanalytic, analytic, and postanalytic considerations for PD-L1 and TMB testing in the clinical setting.

FUTURE DIRECTIONS

PD-L1 is widely recognized as an imperfect biomarker for the selection of patients for ICI therapy. While many patients with NSCLC are now receiving ICI therapy at some stage of their care and recent improvements in cancer outcomes are unparalleled in modern oncologic history, most patients with NSCLC will not derive a response or survival benefit. Beyond lack of response, some patients have severe adverse effects related to aberrant immune activation.[158,159] Analyses of the tumor microenvironment (TME) and host factors have identified other considerations that may influence response to ICI therapy.[1] Relatively simple variables such as $CD8^+$ T-cell enumeration in the TME may add to the predictive power of PD-L1 status, but this approach has not been widely validated in clinical practice.[95,160] More complex analyses, including multiplex immune profiling by IHC or immunofluorescence to capture both the PD-L1/PD-1 status and immune cell characterization in the TME, have been shown to outperform PD-L1 IHC alone. While these strategies have been piloted in a number of academic centers,[161,162] complexities of image analysis and data storage have slowed adoption beyond large academic centers. Other conceptually straightforward analyses such as PD-1/PD-L1 proximity may be superior to PD-L1 status for predicting ICI response, but are not widely available.[163] Host factors such as gut microbiome, human leukocyte antigen (HLA) genotype, and neutrophil to leukocyte ratio have all been examined extensively as factors influencing ICI therapy response and are increasingly incorporated into clinical trials either as components of the primary design or as correlative biomarkers.[164] As highlighted by the widespread interest in TMB as an ICI therapy biomarker, other specific tumor genome characteristics beyond driver oncogene mutation status are subject to extensive study in ICI-treated patient cohorts. Comprehensive characterization of the tumor genome, using next-generation sequencing technology, is required to capture the suite of co-mutations (such as in *serine/threonine kinase 11* [*STK11*], *Kelch-like ECH-associated protein 1* [*KEAP1*], switch/sucrose non-fermentable SWI/SNF pathway genes), mutational signatures (Tobacco, apolipoprotein B mRNA editing enzyme, catalytic polypeptide [APOBEC], microsatellite instability, homologous recombination deficiency), and genome state changes[165] that may inform ICI treatment outcomes.[166] Given the complexity of the tumor-immune interaction, biomarker development is likely to benefit from artificial intelligence algorithms designed to improve the quantification and synthesis of pathologic, genomic, radiomic, and clinical data.[162,167,168] Access to this level of information is currently limited to a small subset of patients with cancer globally. While these types of advanced analyses are essential to drive discovery, it remains of paramount importance to identify easily implemented and cost-effective biomarkers that can identify patients most likely to benefit (or not benefit) from ICI therapy.

CONCLUSIONS

This guideline was developed during the course of 4 years, during which time the use of immunotherapy for patients with lung cancer gained greater traction, and our understanding of the factors that contribute to RRs grew in sophistication. That said, PD-L1 IHC testing remains a cornerstone of NSCLC biomarker testing, and despite its less-than-ideal negative and positive predictive values, most patients with advanced NSCLC will have their tumors tested for PD-L1 expression.[169] The development of different paired diagnostic assays for each of the immunotherapy drugs taken through clinical trials to regulatory approval has created a confusing landscape of companion (on-label "required") diagnostics and complementary (on-label "nice to know") diagnostics, along with a plethora of PD-L1 antibody clones and testing platforms. The EP recognized that the regulatory-approved diagnostics are clinically validated, and as such their use is recommended. However, the panel also recognized that the practical reality of most laboratories—including lack of access to the full suite of approved clones and platforms and the increased cost of running CDx-labeled assays—may require use of LDTs. To ensure patient access to PD-L1 testing, particularly at the local level, this panel also endorses the use of LDTs following technical validation against 1 or more of the approved CDx PD-L1 assays. Formal IHC validation recommendations are beyond the scope of this guideline; the reader is directed to IHC validation guidelines published by the CAP. It is incumbent on testing laboratories to recognize the biological and technical variables that influence PD-L1 expression status, including its expression heterogeneity, the quantitative sample requirements, and appropriate validation of the chosen assay for each of the sample types that may be used to render a diagnosis in patients with lung cancer. It is also important to recognize that other factors may contribute to the decision to proceed with ICI therapy in patients with NSCLC, including the presence of genomic driver alterations such as in *EGFR* and *ALK*, suggesting a lower efficacy of ICI. TMB has been proposed as a pan-cancer biomarker of ICI response, but there is a dearth of published data to date suggesting that the current cut point defining TMB-high is a reliable predictor of response to ICI in patients with NSCLC.

Guideline Revision

This guideline will be assessed every 5 years to determine if an update is warranted. When appropriate, the panel may recommend an earlier update to the CAP in collaboration with ASCO, AMP, IASLC, PPS, and LUNGevity Foundation.

Disclaimer

The CAP developed the Pathology and Laboratory Quality Center for Evidence-Based Guidelines as a forum to create and maintain laboratory practice guidelines (LPGs). Guidelines are intended to assist physicians and patients in clinical decision-making and to identify questions and settings for further research. With the rapid flow of scientific information, new evidence may emerge between the time an LPG is developed and when it is published or read. LPGs are not continually updated and may not reflect the most recent evidence. LPGs address only the topics specifically identified

therein and are not applicable to other interventions, diseases, or stages of diseases. Furthermore, guidelines cannot account for individual variation among patients and cannot be considered inclusive of all proper methods of care or exclusive of other treatments. It is the responsibility of the treating physician or other health care provider, relying on independent experience and knowledge, to determine the best course of treatment for the patient. Accordingly, adherence to any LPG is voluntary, with the ultimate determination regarding its application to be made by the physician in light of each patient's individual circumstances and preferences. CAP makes no warranty, expressed or implied, regarding LPGs and specifically excludes any warranties of merchantability and fitness for a particular use or purpose. CAP assumes no responsibility for any injury or damage to persons or property arising out of or related to any use of this statement or for any errors or omissions.

The authors thank the collaborating societies and their staff involved in the development of this guideline: American Society of Clinical Oncology, Association for Molecular Pathology, International Association for the Study of Lung Cancer, Pulmonary Pathology Society, and LUNGevity Foundation. The authors also gratefully acknowledge advisory panel members for their careful review and guidance throughout the development of the guideline and for their thoughtful review of this work: Ezra Baraban, MD, Eric Bernicker, MD, Russell Broaddus, MD, PhD, Sanja Dacic, MD, PhD, Fang Fan, MD, PhD, Patrick Fitzgibbons, MD, Zaibo Li, MD, PhD, Robert McGee, MD, PhD, Sinchita Roy-Chowdhuri, MD, PhD, Marina Vivero, MD, Barbara A. Ward, BA, Ahmet Zehir, PhD; and Carol F. Colasacco, AHIP, MLIS, SCT(ASCP), Nicole Thomas, MPH, CT(ASCP), and Marisol Hernandez, MLS, MA for their support throughout the guideline development process.

References

1. Mino-Kenudson M, Schalper K, Cooper W, et al. Predictive Biomarkers for Immunotherapy in Lung Cancer: Perspective From the International Association for the Study of Lung Cancer Pathology Committee. *J Thorac Oncol*. 2022; 17(12):1335–1354. doi:10.1016/j.jtho.2022.09.109

2. Lantuejoul S, Damiola F, Adam J. Selected highlights of the 2019 Pulmonary Pathology Society Biennial Meeting: PD-L1 test harmonization studies. *Transl Lung Cancer Res*. 2020;9(3):906–916. doi:10.21037/tlcr.2020.03.23

3. Bailey C, Black JRM, Reading JL, et al. Tracking Cancer Evolution through the Disease Course. *Cancer Discov*. 2021;11(4):916–932. doi:10.1158/2159-8290.cd-20-1559

4. Shen X, Wang Y, Jin Y, et al. PD-L1 expression in non-small cell lung cancer: heterogeneity by pathologic types, tissue sampling and metastasis. *J Thorac Dis*. 2021;13(7):4360–4370. doi:10.21037/jtd-21-388

5. Hwang DM, Albaqer T, Santiago RC, et al. Prevalence and Heterogeneity of PD-L1 Expression by 22C3 Assay in Routine Population-Based and Reflexive Clinical Testing in Lung Cancer. *J Thorac Oncol*. 2021;16(9):1490–1500. doi:10.1016/j.jtho.2021.03.028

6. Institute of Medicine Committee on Standards for Developing Trustworthy Clinical Practice Guidelines. In: Graham R, Mancher M, Miller Wolman D, Greenfield S, Steinberg E, eds. *Clinical Practice Guidelines We Can Trust*. Washington, DC: National Academies Press (US); 2011.

7. College of American Pathologists. Evidence-Based Guideline Development Methodology Manual: Pathology and Laboratory Quality Center for Evidence-Based Guidelines. 2020. https://documents.cap.org/documents/cap-center-ebg-development-manual.pdf?_gl=1*jtqlw5*_ga*ODE2MjE4MzM2LjE2NDc1MzA1NjI.*_ga_97ZFJSQQ0X*MTcwODk1NzcyOC42Ny4xLjE3MDg5NTc3NzYuMC4wLjA. Accessed November 7, 2023.

8. Schuenemann H, Brozek J, Guyatt G, Oxman A. *Handbook for Grading the Quality of Evidence and the Strength of Recommendations Using the GRADE Approach*. 2013.

9. Alonso-Coello P, Schünemann HJ, Moberg J, et al. GRADE Evidence to Decision (EtD) frameworks: a systematic and transparent approach to making well informed healthcare choices—1: Introduction. *BMJ*. 2016;353:i2016–i2016. doi:10.1136/bmj.i2016

10. Cao R, Ma J-T, Zhang S-L, et al. Rational application of the first-line chemotherapy and immune checkpoint inhibitors in advanced nonsmall cell lung cancer: a meta-analysis. *Cancer Med*. 2019;8(11):5033–5046. doi:10.1002/cam4.2407

11. Kim R, Keam B, Hahn S, et al. First-line pembrolizumab versus pembrolizumab plus chemotherapy versus chemotherapy alone in non-small-cell lung cancer: a systematic review and network meta-analysis. *Clin Lung Cancer*. 2019; 20(5):331–338.e4. doi:10.1016/j.cllc.2019.05.009

12. Borghaei H, Paz-Ares L, Horn L, et al. Nivolumab versus docetaxel in advanced nonsquamous non-small-cell lung cancer. *N Engl J Med*. 2015; 373(17):1627–1639. doi:10.1056/NEJMoa1507643

13. Brahmer J, Reckamp KL, Baas P, et al. Nivolumab versus docetaxel in advanced squamous-cell non-small-cell lung cancer. *N Engl J Med*. 2015;373(2): 123–135. doi:10.1056/NEJMoa1504627

14. Fehrenbacher L, Spira A, Ballinger M, et al. Atezolizumab versus docetaxel for patients with previously treated non-small-cell lung cancer (POPLAR): a multicentre, open-label, phase 2 randomised controlled trial. *Lancet*. 2016; 387(10030):1837–1846. doi:10.1016/S0140-6736(16)00587-0

15. Garon EB, Hellmann MD, Rizvi NA, et al. Five-year overall survival for patients with advanced non–small-cell lung cancer treated with pembrolizumab: results from the phase I KEYNOTE-001 study. *J Clin Oncol*. 2019;37(28):2518–2527. doi:10.1200/JCO.19.00934

16. Hellmann MD, Paz-Ares L, Bernabe Caro R, et al. Nivolumab plus ipilimumab in advanced non-small-cell lung cancer. *N Engl J Med*. 2019;381(21): 2020–2031. doi:10.1056/NEJMoa1910231

17. Mok TSK, Wu YL, Kudaba I, et al. Pembrolizumab versus chemotherapy for previously untreated, PD-L1-expressing, locally advanced or metastatic non-small-cell lung cancer (KEYNOTE-042): a randomised, open-label, controlled, phase 3 trial. *Lancet*. 2019;393(10183):1819–1830. doi:10.1016/S0140-6736(18)32409-7

18. Ready N, Hellmann MD, Awad MM, et al. First-line nivolumab plus ipilimumab in advanced non-small-cell lung cancer (CheckMate 568): outcomes by programmed death ligand 1 and tumor mutational burden as biomarkers. *J Clin Oncol*. 2019;37(12):992–1000. doi:10.1200/JCO.18.01042

19. Rittmeyer A, Barlesi F, Waterkamp D, et al. Atezolizumab versus docetaxel in patients with previously treated non-small-cell lung cancer (OAK): a phase 3, open-label, multicentre randomised controlled trial. *Lancet*. 2017; 389(10066):255–265. doi:10.1016/S0140-6736(16)32517-X

20. West H, McCleod M, Hussein M, et al. Atezolizumab in combination with carboplatin plus nab-paclitaxel chemotherapy compared with chemotherapy alone as first-line treatment for metastatic non-squamous non-small-cell lung cancer (IMpower130): a multicentre, randomised, open-label, phase 3 trial. *Lancet Oncol*. 2019;20(7):924–937. doi:10.1016/S1470-2045(19)30167-6

21. Garon EB, Rizvi NA, Hui R, et al. Pembrolizumab for the treatment of non-small-cell lung cancer. *N Engl J Med*. 2015;372(21):2018–2028. doi:10.1056/NEJMoa1501824

22. Herbst RS, Baas P, Perez-Gracia JL, et al. Use of archival versus newly collected tumor samples for assessing PD-L1 expression and overall survival: an updated analysis of keynote-010 trial. *Ann Oncol*. 2019;30(2):281–289. doi:10.1093/annonc/mdy545

23. Horn L, Gettinger SN, Gordon MS, et al. Safety and clinical activity of atezolizumab monotherapy in metastatic non-small-cell lung cancer: final results from a phase I study. *Eur J Cancer*. 2018;101:201–209. doi:10.1016/j.ejca.2018.06.031

24. Hui R, Garon EB, Goldman JW, et al. Pembrolizumab as first-line therapy for patients with PD-L1-positive advanced non-small cell lung cancer: a phase 1 trial. *Ann Oncol*. 2017;28(4):874–881. doi:10.1093/annonc/mdx008

25. Antonia SJ, Balmanoukian A, Brahmer J, et al. Clinical activity, tolerability, and long-term follow-up of durvalumab in patients with advanced NSCLC. *J Thorac Oncol*. 2019;14(10):1794–1806. doi:10.1016/j.jtho.2019.06.010

26. Gettinger S, Horn L, Jackman D, et al. Five-year follow-up of nivolumab in previously treated advanced non–small-cell lung cancer: results from the CA209-003 study. *J Clin Oncol*. 2018;36(17):1675–1684. doi:10.1200/JCO.2017.77.0412

27. Gettinger S, Rizvi NA, Chow LQ, et al. Nivolumab monotherapy for first--line treatment of advanced non-small-cell lung cancer. *J Clin Oncol*. 2016; 34(25):2980–2987. doi:10.1200/JCO.2016.66.9929

28. Peters S, Gettinger S, Johnson ML, et al. Phase II trial of atezolizumab as first-line or subsequent therapy for patients with programmed death-ligand 1-selected advanced non-small-cell lung cancer (BIRCH). *J Clin Oncol*. 2017; 35(24):2781–2789. doi:10.1200/JCO.2016.71.9476

29. Aguilar EJ, Ricciuti B, Gainor JF, et al. Outcomes to first-line pembrolizumab in patients with non-small cell lung cancer and very high PD-L1 expression. *Ann Oncol*. 2019;30(10):1653–1659. doi:10.1093/annonc/mdz288

30. Ahn BC, Pyo KH, Xin CF, et al. Comprehensive analysis of the characteristics and treatment outcomes of patients with non-small cell lung cancer treated with anti-PD-1 therapy in real-world practice. *J Cancer Res Clin Oncol*. 2019; 145(6):1613–1323. doi:10.1007/s00432-019-02899-y

31. Edahiro R, Kanazu M, Kurebe H, et al. Clinical outcomes in non-small cell lung cancer patients with an ultra-high expression of programmed death ligand-1 treated using pembrolizumab as a first-line therapy: a retrospective multicenter cohort study in Japan. *PLoS One*. 2019;14(7). doi:10.1371/journal.pone.0220570

32. Kim HS, Cha H, Kim J, et al. Genomic scoring to determine clinical benefit of immunotherapy by targeted sequencing. *Eur J Cancer*. 2019;120:65–74. doi:10.1016/j.ejca.2019.08.001

33. Lin SY, Yang CY, Liao BC, et al. Tumor PD-L1 expression and clinical outcomes in advanced-stage non-small cell lung cancer patients treated with nivolumab or pembrolizumab: real-world data in Taiwan. *J Cancer*. 2018;9(10):1813–1820. doi:10.7150/jca.24985

34. Oya Y, Yoshida T, Kuroda H, et al. Predictive clinical parameters for the response of nivolumab in pretreated advanced non-small-cell lung cancer. *Oncotarget*. 2017;8(61):103117–103128. doi:10.18632/oncotarget.21602

35. Rizvi H, Sanchez-Vega F, La K, et al. Molecular determinants of response to anti-programmed cell death (PD)-1 and anti-programmed death-ligand 1 (PD-L1) blockade in patients with non-small-cell lung cancer profiled with

targeted next-generation sequencing. *J Clin Oncol*. 2018;36(7):633–641. doi:10.1200/JCO.2017.75.3384

36. Tamiya M, Tamiya A, Hosoya K, et al. Efficacy and safety of pembrolizumab as first-line therapy in advanced non-small cell lung cancer with at least 50% PD-L1 positivity: a multicenter retrospective cohort study (HOPE-001). *Invest New Drugs*. 2019;37(6):1266–1273. doi:10.1007/s10637-019-00843-y

37. Fujimoto D, Sato Y, Uehara K, et al. Predictive performance of four programmed cell death ligand 1 assay systems on nivolumab response in previously treated patients with non-small cell lung cancer. *J Thorac Oncol*. 2018;13(3):377–386. doi:10.1016/j.jtho.2017.11.123

38. Tseng J-S, Yang T-Y, Wu C-Y, et al. Characteristics and predictive value of PD-L1 status in real-world non-small cell lung cancer patients. *J Immunother*. 2018;41(6):292–299. doi:10.1097/CJI.0000000000000226

39. Reck M, Rodriguez-Abreu D, Robinson AG, et al. Five-year outcomes with pembrolizumab versus chemotherapy for metastatic non-small-cell lung cancer with PD-L1 tumor proportion score >= 50. *J Clin Oncol*. 2021;39(21):2339–2234. doi:10.1200/JCO.21.00174

40. Park K, Ozguroglu M, Vansteenkiste J, et al. Avelumab versus docetaxel in patients with platinum-treated advanced NSCLC: 2-year follow-up from the JAVELIN Lung 200 phase 3 trial. *J Thorac Oncol*. 2021;16(8):1369–1378. doi:10.1016/j.jtho.2021.03.009

41. Herbst RS, Garon EB, Kim DW, et al. Five year survival update from KEYNOTE-010: pembrolizumab versus docetaxel for previously treated, programmed death-ligand 1-positive advanced NSCLC. *J Thorac Oncol*. 2021;16(10):1718–1732. doi:10.1016/j.jtho.2021.05.001

42. Paz-Ares LG, Ramalingam SS, Ciuleanu TE, et al. First-line nivolumab plus ipilimumab in advanced NSCLC: 4-year outcomes from the randomized, open-label, phase 3 CheckMate 227 part 1 trial. *J Thorac Oncol*. 2022;17(2):289–308. doi:10.1016/j.jtho.2021.09.010

43. Jassem J, de Marinis F, Giaccone G, et al. Updated overall survival analysis from IMpower110: atezolizumab versus platinum-based chemotherapy in treatment-naive programmed death-ligand 1-selected NSCLC. *J Thorac Oncol*. 2021;16(11):1872–1882. doi:10.1016/j.jtho.2021.06.019

44. Felip E, Altorki N, Zhou C, et al. Adjuvant atezolizumab after adjuvant chemotherapy in resected stage IB-IIIA non-small-cell lung cancer (IMpower010): a randomised, multicentre, open-label, phase 3 trial. *Lancet*. 2021;398(10308):1344–1357. doi:10.1016/S0140-6736(21)02098-5

45. Forde PM, Spicer J, Lu S, et al. Neoadjuvant nivolumab plus chemotherapy in resectable lung cancer. *N Engl J Med*. 2022;386(21):1973–1985. doi:10.1056/NEJMoa2202170

46. Mazieres J, Drilon A, Lusque A, et al. Immune checkpoint inhibitors for patients with advanced lung cancer and oncogenic driver alterations: results from the IMMUNOTARGET registry. *Ann Oncol*. 2019;30(8):1321–1328. doi:10.1093/annonc/mdz167

47. Keytruda [EPAR product information]. Merck Sharp & Dohme BV; 2015.

48. Oshima Y, Tanimoto T, Yuji K, Tojo A. EGFR-TKI-associated interstitial pneumonitis in nivolumab-treated patients with non-small cell lung cancer. *JAMA Oncol*. 2018;4(8):1112–1115. doi:10.1001/jamaoncol.2017.4526

49. Schoenfeld AJ, Arbour KC, Rizvi H, et al. Severe immune-related adverse events are common with sequential PD-(L)1 blockade and osimertinib. *Ann Oncol*. 2019;30(5):839–844. doi:10.1093/annonc/mdz077

50. Forest F, Cote G, Laville D, et al. Impact of delayed fixation and decalcification on PD-L1 expression: a comparison of two clones. *Virchows Arch*. 2019;475(6):693–699. doi:10.1007/s00428-019-02613-w

51. Ng TL, Liu Y, Dimou A, et al. Predictive value of oncogenic driver subtype, programmed death-1 ligand (PD-L1) score, and smoking status on the efficacy of PD-1/PD-L1 inhibitors in patients with oncogene-driven non-small cell lung cancer. *Cancer*. 2019;125(7):1038–1049. doi:10.1002/cncr.31871

52. Griffith SD, Tucker M, Bowser B, et al. Generating real-world tumor burden endpoints from electronic health record data: comparison of RECIST, radiology-anchored, and clinician-anchored approaches for abstracting real-world progression in non-small cell lung cancer. *Adv Ther*. 2019;36(8):2122–2136. doi:10.1007/s12325-019-00970-1

53. Planchard D, Besse B, Groen HJM, et al. Phase 2 study of dabrafenib plus trametinib in patients with BRAF V600E-mutant metastatic NSCLC: updated 5-year survival rates and genomic analysis. *J Thorac Oncol*. 2022;17(1):103–115. doi:10.1016/j.jtho.2021.08.011

54. Wolf J, Seto T, Han JY, et al. Capmatinib in MET exon 14-mutated or MET-amplified non-small-cell lung cancer. *N Engl J Med*. 2020;383(10):944–957. doi:10.1056/NEJMoa2002787

55. Drilon A, Oxnard GR, Tan DSW, et al. Efficacy of selpercatinib in RET fusion-positive non-small-cell lung cancer. *N Engl J Med*. 2020;383(9):813–824. doi:10.1056/NEJMoa2005653

56. Lindeman NI, Cagle PT, Aisner DL, et al. Updated Molecular Testing Guideline for the Selection of Lung Cancer Patients for Treatment With Targeted Tyrosine Kinase Inhibitors: guideline from the College of American Pathologists, the International Association for the Study of Lung Cancer, and the Association for Molecular Pathology. *Arch Pathol Lab Med*. 2018;142(3):321–346. doi:10.5858/arpa.2017-0388-CP

57. Fitzgibbons PL, Bradley LA, Fatheree LA, et al. Principles of analytic validation of immunohistochemical assays: guideline from the College of American Pathologists Pathology and Laboratory Quality Center. *Arch Pathol Lab Med*. 2014;138(11):1432–1443. doi:10.5858/arpa.2013-0610-CP

58. Torous VF, Rangachari D, Gallant BP, Shea M, Costa DB, VanderLaan PA. PD-L1 testing using the clone 22C3 pharmDx kit for selection of patients with non-small cell lung cancer to receive immune checkpoint inhibitor therapy: are cytology cell blocks a viable option? *J Am Soc Cytopathol*. 2018;7(3):133–141. doi:10.1016/j.jasc.2018.02.003

59. Keller MD, Neppl C, Irmak Y, et al. Adverse prognostic value of PD-L1 expression in primary resected pulmonary squamous cell carcinomas and paired mediastinal lymph node metastases. *Mod Pathol*. 2018;31(1):101–110. doi:10.1038/modpathol.2017.111

60. Kim HR, Cha YJ, Hong MH, et al. Concordance of programmed death-ligand 1 expression between primary and metastatic non-small cell lung cancer by immunohistochemistry and RNA in situ hybridization. *Oncotarget*. 2017;8(50):87234–87243. doi:10.18632/oncotarget.20254

61. Kim S, Koh J, Kwon D, et al. Comparative analysis of PD-L1 expression between primary and metastatic pulmonary adenocarcinomas. *Eur J Cancer*. 2017;75:141–149. doi:10.1016/j.ejca.2017.01.004

62. Sheffield BS, Fulton R, Kalloger SE, et al. Investigation of PD-L1 biomarker testing methods for PD-1 axis inhibition in non-squamous non–small cell lung cancer. *J Histochem Cytochem*. 2016;64(10):587–600. doi:10.1369/0022155416665338

63. Téglási V, Pipek O, Lózsa R, et al. PD-L1 expression of lung cancer cells, unlike infiltrating immune cells, is stable and unaffected by therapy during brain metastasis. *Clin Lung Cancer*. 2019;20(5):363–369.e2. doi:10.1016/j.cllc.2019.05.008

64. Uruga H, Bozkurtlar E, Huynh TG, et al. Programmed cell death ligand (PD-L1) expression in stage II and III lung adenocarcinomas and nodal metastases. *J Thorac Oncol*. 2017;12(3):458–466. doi:10.1016/j.jtho.2016.10.015

65. Munari E, Zamboni G, Lunardi G, et al. PD-L1 expression comparison between primary and relapsed non-small cell lung carcinoma using whole sections and clone SP263. *Oncotarget*. 2018;9(54):30465–30471. doi:10.18632/oncotarget.25770

66. Wang H, Agulnik J, Kasymjanova G, et al. The metastatic site does not influence PD-L1 expression in advanced non-small cell lung carcinoma. *Lung Cancer*. 2019;132:36–38. doi:10.1016/j.lungcan.2019.04.009

67. Grosu HB, Arriola A, Stewart J, et al. PD-L1 detection in histology specimens and matched pleural fluid cell blocks of patients with NSCLC. *Respirology*. 2019;24(12):1198–1203. doi:10.1111/resp.13614

68. Hernandez A, Brandler TC, Zhou F, Moreira AL, Schatz-Siemers N, Simsir A. Assessment of programmed death–ligand 1 (PD-L1) immunohistochemical expression on cytology specimens in non–small cell lung carcinoma: a comparative study with paired surgical specimens. *Am J Clin Pathol*. 2019;151(4):403–415. doi:10.1093/AJCP/AQY164

69. Kuempers C, van der Linde LIS, Reischl M, et al. Comparison of PD-L1 expression between paired cytologic and histologic specimens from non-small cell lung cancer patients. *Virchows Arch*. 2020;476(2):261–271. doi:10.1007/s00428-019-02632-7

70. Mei P, Shilo K, Wei L, Shen R, Tonkovich D, Li Z. Programmed cell death ligand 1 expression in cytologic and surgical non–small cell lung carcinoma specimens from a single institution: association with clinicopathologic features and molecular alterations. *Cancer Cytopathol*. 2019;127(7):447–457. doi:10.1002/cncy.22140

71. Munari E, Zamboni G, Sighele G, et al. Expression of programmed cell death ligand 1 in non–small cell lung cancer: comparison between cytologic smears, core biopsies, and whole sections using the SP263 assay. *Cancer Cytopathol*. 2019;127(1):52–61. doi:10.1002/cncy.22083

72. Noll B, Wang WL, Gong Y, et al. Programmed death ligand 1 testing in non–small cell lung carcinoma cytology cell block and aspirate smear preparations. *Cancer Cytopathol*. 2018;126(5):342–352. doi:10.1002/cncy.21987

73. Ilie M, Juco J, Huang L, Hofman V, Khambata-Ford S, Hofman P. Use of the 22C3 anti-programmed death-ligand 1 antibody to determine programmed death-ligand 1 expression in cytology samples obtained from non-small cell lung cancer patients. *Cancer Cytopathol*. 2018;126(4):264–274. doi:10.1002/cncy.21977

74. Skov BG, Skov T. Paired comparison of PD-L1 expression on cytologic and histologic specimens from malignancies in the lung assessed with PD-L1 IHC 28-8pharmDx and PD-L1 IHC 22C3pharmDx. *Appl Immunohistochem Mol Morphol*. 2017;25(7):453–459. doi:10.1097/PAI.0000000000000540

75. Russell-Goldman E, Kravets S, Dahlberg SE, Sholl LM, Vivero M. Cytologic-histologic correlation of programmed death-ligand 1 immunohistochemistry in lung carcinomas. *Cancer Cytopathol*. 2018;126(4):253–263. doi:10.1002/cncy.21973

76. Daverio M, Patrucco F, Gavelli F, et al. Comparative analysis of programmed death ligand 1 expression in paired cytologic and histologic specimens of non-small cell lung cancer. *Cancer Cytopathol*. 2020;128(8):580–588. doi:10.1002/cncy.22292

77. Lou SK, Ko HM, Kinoshita T, et al. Implementation of PD-L1 22C3 IHC pharmDxTM in cell block preparations of lung cancer: concordance with surgical resections and technical validation of CytoLyt R prefixation. *Acta Cytol*. 2020;64(6):577–587. doi:10.1159/000508628

78. Lozano MD, Abengozar-Muela M, Echeveste JI, et al. Programmed death--ligand 1 expression on direct Pap-stained cytology smears from non-small cell lung cancer: comparison with cell blocks and surgical resection specimens. *Cancer Cytopathol*. 2019;127(7):470–480. doi:10.1002/cncy.22155

79. Song SG, Lee J, Koh J, Kim S, Chung DH, Jeon YK. Utility of PD-L1 immunocytochemistry using body-fluid cell blocks in patients with non-small-cell lung cancer. *Diagn Cytopathol*. 2020;48(4):291–299. doi:10.1002/dc.24379

80. Zou Y, Xu L, Tang Q, et al. Cytology cell blocks from malignant pleural effusion are good candidates for PD-L1 detection in advanced NSCLC compared with matched histology samples. *BMC Cancer*. 2020;20(1):344. doi:10.1186/s12885-020-06851-z

81. Ilie M, Long-Mira E, Bence C, et al. Comparative study of the PD-L1 status between surgically resected specimens and matched biopsies of NSCLC patients

reveal major discordances: a potential issue for anti-PD-L1 therapeutic strategies. *Ann Oncol*. 2016;27(1):147–153. doi:10.1093/annonc/mdv489

82. Elfving H, Mattsson JSM, Lindskog C, Backman M, Menzel U, Micke P. Programmed cell death ligand 1 immunohistochemistry: a concordance study between surgical specimen, biopsy, and tissue microarray. *Clin Lung Cancer*. 2019;20(4):258–262.e1. doi:10.1016/j.cllc.2019.02.012

83. Gradecki SE, Grange JS, Stelow EB. Concordance of PD-L1 expression between core biopsy and resection specimens of non-small cell lung cancer. *Am J Surg Pathol*. 2018;42(8):1090–1094. doi:10.1097/PAS.0000000000001085

84. Sakata KK, Midthun DE, Mullon JJ, et al. Comparison of programmed death ligand-1 immunohistochemical staining between endobronchial ultrasound transbronchial needle aspiration and resected lung cancer specimens. *Chest*. 2018;154(4):827–837. doi:10.1016/j.chest.2018.07.017

85. Wang G, Ionescu DN, Lee CH, et al. PD-L1 testing on the EBUS-FNA cytology specimens of non-small cell lung cancer. *Lung Cancer*. 2019;136:1–5. doi:10.1016/j.lungcan.2019.07.033

86. Vigliar E, Malapelle U, Iaccarino A, et al. PD-L1 expression on routine samples of non-small cell lung cancer: results and critical issues from a 1-year experience of a centralised laboratory. *J Clin Pathol*. 2019;72(6):412–417. doi:10.1136/jclinpath-2019-205732

87. Wang H, Agulnik J, Kasymjanova G, et al. Cytology cell blocks are suitable for immunohistochemical testing for PD-L1 in lung cancer. *Ann Oncol*. 2018;29(6):1417–1422. doi:10.1093/annonc/mdy126

88. Chan AWH, Tong JHM, Kwan JSH, et al. Assessment of programmed cell death ligand-1 expression by 4 diagnostic assays and its clinicopathological correlation in a large cohort of surgical resected non-small cell lung carcinoma. *Mod Pathol*. 2018;31(9):1381–1390. doi:10.1038/s41379-018-0053-3

89. Munari E, Zamboni G, Lunardi G, et al. PD-L1 expression heterogeneity in non–small cell lung cancer: defining criteria for harmonization between biopsy specimens and whole sections. *J Thorac Oncol*. 2018;13(8):1113–1120. doi:10.1016/j.jtho.2018.04.017

90. Rehman JA, Han G, Carvajal-Hausdorf DE, et al. Quantitative and pathologist-read comparison of the heterogeneity of programmed death-ligand 1 (PD-L1) expression in non-small cell lung cancer. *Mod Pathol*. 2017;30(3):340–349. doi:10.1038/modpathol.2016.186

91. Hernandez A, Brandler TC, Chen F, et al. Scoring of programmed death-ligand 1 immunohistochemistry on cytology cell block specimens in non-small cell lung carcinoma. *Am J Clin Pathol*. 2020;154(4):517–524. doi:10.1093/ajcp/aqaa073

92. Agilent. *PD-L IHC 22C3 pharmDx: Interpretation Manual—NSCLC*; Agilent Technologies; 2021.

93. Gagné A, Wang E, Bastien N, et al. Impact of specimen characteristics on PD-L1 testing in non-small cell lung cancer: validation of the IASLC PD-L1 testing guidelines. *J Thorac Oncol*. 2019;14(12):2062–2070. doi:10.1016/j.jtho.2019.08.2503

94. Naito T, Udagawa H, Sato J, et al. A minimum of 100 tumor cells in a single biopsy sample is required to assess programmed cell death ligand 1 expression in predicting patient response to nivolumab treatment in nonsquamous non–small cell lung carcinoma. *J Thorac Oncol*. 2019;14(10):1818–1827. doi:10.1016/j.jtho.2019.06.019

95. Conde E, Caminoa A, Dominguez C, et al. Aligning digital CD8(+) scoring and targeted next-generation sequencing with programmed death ligand 1 expression: a pragmatic approach in early-stage squamous cell lung carcinoma. *Histopathology*. 2018;72(2):270–284. doi:10.1111/his.13346

96. Casadevall D, Clave S, Taus A, et al. Heterogeneity of tumor and immune cell PD-L1 expression and lymphocyte counts in surgical NSCLC samples. *Clin Lung Cancer*. 2017;18(6):682–691 e5. doi:10.1016/j.cllc.2017.04.014

97. Dong ZY, Zhang C, Li YF, et al. Genetic and immune profiles of solid predominant lung adenocarcinoma reveal potential immunotherapeutic strategies. *J Thorac Oncol*. 2018;13(1):85–96. doi:10.1016/j.jtho.2017.10.020

98. Bigras G, Mairs S, Swanson PE, Morel D, Lai R, Izevbaye I. Small biopsies misclassify up to 35% of PD-L1 assessments in advanced lung non-small cell lung carcinomas. *Appl Immunohistochem Mol Morphol*. 2018;26(10):701–708. doi:10.1097/pai.0000000000000698

99. Bubendorf L, Conde E, Cappuzzo F, et al. A noninterventional, multinational study to assess PD-L1 expression in cytological and histological lung cancer specimens. *Cancer Cytopathol*. 2020;128(12):928–938. doi:10.1002/cncy.22324

100. Grillo F, Pigozzi S, Ceriolo P, Calamaro P, Fiocca R, Mastracci L. Factors affecting immunoreactivity in long-term storage of formalin-fixed paraffin-embedded tissue sections. *Histochem Cell Biol*. 2015;144(1):93–99. doi:10.1007/s00418-015-1316-4

101. Grillo F, Bruzzone M, Pigozzi S, et al. Immunohistochemistry on old archival paraffin blocks: is there an expiry date? *J Clin Pathol*. 2017;70(11):988–993. doi:10.1136/jclinpath-2017-204387

102. Fernandez AI, Gaule P, Rimm DL. Tissue age affects antigenicity and scoring for the 22C3 immunohistochemistry companion diagnostic test. *Mod Pathol*. 2023;36(7):100159. doi:10.1016/j.modpat.2023.100159

103. Sheng J, Fang W, Yu J, et al. Expression of programmed death ligand-1 on tumor cells varies pre and post chemotherapy in non-small cell lung cancer. *Sci Rep*. 2016;6:20090. doi:10.1038/srep20090

104. Herbst RS, Baas P, Kim DW, et al. Pembrolizumab versus docetaxel for previously treated, PD-L1-positive, advanced non-small-cell lung cancer (KEYNOTE-010): a randomised controlled trial. *Lancet*. 2016;387(10027):1540–1550. doi:10.1016/S0140-6736(15)01281-7

105. Strickland AL, Blacketer S, Molberg K, Markantonis J, Lucas E. Effects of decalcifying agents of variable duration on PD-L1 immunohistochemistry. *Am J Clin Pathol*. 2020;153(2):258–265. doi:10.1093/ajcp/aqz161

106. Pontarollo G, Confavreux CB, Pialat JB, et al. Bone decalcification to assess programmed cell death ligand 1 expression in bone metastases of non-small cell lung cancers. *J Bone Oncol*. 2020;21:100275. doi:10.1016/j.jbo.2020.100275

107. Krawczyk P, Jarosz B, Kucharczyk T, et al. Immunohistochemical assays incorporating SP142 and 22C3 monoclonal antibodies for detection of PD-L1 expression in NSCLC patients with known status of EGFR and ALK genes. *Oncotarget*. 2017;8(38):64283–64293. doi:10.18632/oncotarget.19724

108. Xu H, Lin G, Huang C, et al. Assessment of concordance between 22C3 and SP142 immunohistochemistry assays regarding PD-L1 expression in non-small cell lung cancer. *Sci Rep*. 2017;7(1):16956. doi:10.1038/s41598-017-17034-5

109. Saito T, Tsuta K, Ishida M, et al. Comparative study of programmed cell death ligand-1 immunohistochemistry assays using 22C3 and 28-8 antibodies for non-small cell lung cancer: analysis of 420 surgical specimens from Japanese patients. *Lung Cancer*. 2018;125:230–237. doi:10.1016/j.lungcan.2018.10.005

110. Beck KS, Kim SJ, Kang JH, Han DH, Jung JI, Lee KY. CT-guided transthoracic needle biopsy for evaluation of PD-L1 expression: comparison of 22C3 and SP263 assays. *Thorac Cancer*. 2019;10(7):1612–1618. doi:10.1111/1759-7714.13126

111. Song P, Guo L, Li W, Zhang F, Ying J, Gao S. Clinicopathologic correlation with expression of PD-L1 on both tumor cells and tumor-infiltrating immune cells in patients with non-small cell lung cancer. *J Immunother*. 2019;42(1):23–28. doi:10.1097/CJI.0000000000000249

112. Fujimoto D, Yamashita D, Fukuoka J, et al. Comparison of PD-L1 assays in non-small cell lung cancer: 22C3 pharmDx and SP263. *Anticancer Res*. 2018;38(12):6891–6895. doi:10.21873/anticanres.13065

113. Humphries MP, McQuaid S, Craig SG, et al. Critical appraisal of programmed death ligand 1 reflex diagnostic testing: current standards and future opportunities. *J Thorac Oncol*. 2019;14(1):45–53. doi:10.1016/j.jtho.2018.09.025

114. Park HY, Oh IJ, Kho BG, et al. Clinical characteristics of Korean patients with lung cancer who have programmed death-ligand 1 expression. *Tuberc Respir Dis*. 2019;82(3):227–233. doi:10.4046/trd.2018.0070

115. Hirsch FR, McElhinny A, Stanforth D, et al. PD-L1 Immunohistochemistry assays for lung cancer: results from phase 1 of the Blueprint PD-L1 IHC Assay Comparison Project. *J Thorac Oncol*. 2017;12(2):208–222. doi:10.1016/j.jtho.2016.11.2228

116. Ratcliffe MJ, Sharpe A, Midha A, et al. Agreement between programmed cell death ligand-1 diagnostic assays across multiple protein expression cutoffs in non-small cell lung cancer. *Clin Cancer Res*. 2017;23(14):3585–3591. doi:10.1158/1078-0432.CCR-16-2375

117. Pang C, Yin L, Zhou X, et al. Assessment of programmed cell death ligand-1 expression with multiple immunohistochemistry antibody clones in non-small cell lung cancer. *J Thorac Dis*. 2018;10(2):816–824. doi:10.21037/jtd.2018.01.124

118. Yeo MK, Choi SY, Seong IO, Suh KS, Kim JM, Kim KH. Association of PD-L1 expression and PD-L1 gene polymorphism with poor prognosis in lung adenocarcinoma and squamous cell carcinoma. *Hum Pathol*. 2017;68:103–111. doi:10.1016/j.humpath.2017.08.016

119. Herbst RS, Giaccone G, de Marinis F, et al. Atezolizumab for first-line treatment of PD-L1-selected patients with NSCLC. *N Engl J Med*. 2020;383(14):1328–1339. doi:10.1056/NEJMoa1917346

120. Parra ER, Villalobos P, Mino B, Rodriguez-Canales J. Comparison of different antibody clones for immunohistochemistry detection of programmed cell death ligand 1 (PD-L1) on non-small cell lung carcinoma. *Appl Immunohistochem Mol Morphol*. 2018;26(2):83–93. doi:10.1097/PAI.0000000000000531

121. Brunnström H, Johansson A, Westbom-Fremer S, et al. PD-L1 immunohistochemistry in clinical diagnostics of lung cancer: Inter-pathologist variability is higher than assay variability. *Mod Pathol*. 2017;30(10):1411–1421. doi:10.1038/modpathol.2017.59

122. Cooper WA, Russell PA, Cherian M, et al. Intra- and interobserver reproducibility assessment of PD-L1 biomarker in non–small cell lung cancer. *Clin Cancer Res*. 2017;23(16):4569–4577. doi:10.1158/1078-0432.CCR-17-0151

123. Marchetti A, Barberis M, Franco R, et al. Multicenter comparison of 22C3 PharmDx (Agilent) and SP263 (Ventana) assays to test PD-L1 expression for NSCLC patients to be treated with immune checkpoint inhibitors. *J Thorac Oncol*. 2017;12(11):1654–1663. doi:10.1016/j.jtho.2017.07.031

124. Munari E, Zamboni G, Lunardi G, et al. PD-L1 expression in non-small cell lung cancer: evaluation of the diagnostic accuracy of a laboratory-developed test using clone E1L3N in comparison with 22C3 and SP263 assays. *Hum Pathol*. 2019;90:54–59. doi:10.1016/j.humpath.2019.05.003

125. Rimm DL, Han G, Taube JM, et al. A prospective, multi-institutional, pathologist-based assessment of 4 immunohistochemistry assays for PD-L1 expression in non-small cell lung cancer. *JAMA Oncol*. 2017;3(8):1051–1058. doi:10.1001/jamaoncol.2017.0013

126. Tsao MS, Kerr KM, Kockx M, et al. PD-L1 immunohistochemistry comparability study in real-life clinical samples: results of Blueprint Phase 2 Project. *J Thorac Oncol*. 2018;13(9):1302–1311. doi:10.1016/j.jtho.2018.05.013

127. Barlesi F, Vansteenkiste J, Spigel D, et al. Avelumab versus docetaxel in patients with platinum-treated advanced non-small-cell lung cancer (JAVELIN Lung 200): an open-label, randomised, phase 3 study. *Lancet Oncol*. 2018;19(11):1468–1479. doi:10.1016/S1470-2045(18)30673-9

128. Lantuejoul S, Sound-Tsao M, Cooper WA, et al. PD-L1 testing for lung cancer in 2019: perspective from the IASLC Pathology Committee. *J Thorac Oncol*. 2020;15(4):499–519. doi:10.1016/j.jtho.2019.12.107

129. Mino-Kenudson M, Le Stang N, Daigneault JB, et al. The International Association for the Study of Lung Cancer Global Survey on Programmed Death-Ligand 1 Testing for NSCLC. *J Thorac Oncol*. 2021;16(4):686–696. doi:10.1016/j.jtho.2020.12.026

130. US Food and Drug Administration. FDA approves cemiplimab-rwlc for non--small cell lung cancer with high PD-L1 expression. https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-cemiplimab-rwlc-non-small-cell-lung-cancer-high-pd-l1-expression. Published February 22, 2021. Accessed September 1, 2023.

131. Grote HJ, Feng Z, Schlichting M, et al. Programmed death-ligand 1 immunohistochemistry assay comparison studies in NSCLC: characterization of the 73-10 assay. *J Thorac Oncol*. 2020;15(8):1306–1316. doi:10.1016/j.jtho.2020.04.013

132. Munari E, Rossi G, Zamboni G, et al. PD-L1 assays 22C3 and SP263 are not interchangeable in non-small cell lung cancer when considering clinically relevant cutoffs: an interclone evaluation by differently trained pathologists. *Am J Surg Pathol*. 2018;42(10):1384–1389. doi:10.1097/PAS.0000000000001105

133. Villaruz LC, Ancevski Hunter K, Kurland BF, Abberbock S, Herbst C, Dacic S. Comparison of PD-L1 immunohistochemistry assays and response to PD-1/L1 inhibitors in advanced non-small-cell lung cancer in clinical practice. *Histopathology*. 2019;74(2):269–275. doi:10.1111/his.13729

134. Adam J, Le Stang N, Rouquette I, et al. Multicenter harmonization study for PD-L1 IHC testing in non-small-cell lung cancer. *Ann Oncol*. 2018;29(4):953–958. doi:10.1093/annonc/mdy014

135. Kim H, Kwon HJ, Park SY, Park E, Chung J-H. PD-L1 immunohistochemical assays for assessment of therapeutic strategies involving immune checkpoint inhibitors in non-small cell lung cancer: a comparative study. *Oncotarget*. 2017;8(58):98524–98532. doi:10.18632/oncotarget.21567

136. Sughayer MA, Alnaimy F, Alsughayer AM, Qamhia N. Comparison of 22C3 PharmDx and SP263 assays to test PD-L1 expression in NSCLC. *Appl Immunohistochem Mol Morphol*. 2019;27(9):663–666. doi:10.1097/PAI.0000000000000671

137. Centers for Medicare & Medicaid Services. LDT and CLIA FAQs. https://www.cms.gov/regulations-and-guidance/legislation/clia/downloads/ldt-and-clia_faqs.pdf. Published October 22, 2013. Accessed November 7, 2023.

138. Sarata A. FDA regulation of laboratory-developed tests (LDTs). Congressional Research Service. https://crsreports.congress.gov/product/pdf/IF/IF11389. Updated December 7, 2022. Accessed November 7, 2023.

139. US Food and Drug Administration. Laboratory-developed tests. https://www.fda.gov/medical-devices/in-vitro-diagnostics/laboratory-developed-tests. Updated September 27, 2018. Accessed November 7, 2023.

140. Buttner R, Gosney JR, Skov BG, et al. Programmed death-ligand 1 immunohistochemistry testing: a review of analytical assays and clinical implementation in non-small-cell lung cancer. *J Clin Oncol*. 2017;35(34):3867–3876. doi:10.1200/JCO.2017.74.7642

141. Scheel AH, Baenfer G, Baretton G, et al. Interlaboratory concordance of PD-L1 immunohistochemistry for non-small-cell lung cancer. *Histopathology*. 2018;72(3):449–459. doi:10.1111/his.13375

142. Centers for Medicare & Medicaid Services. Standard: establishment and verification of performance specifications. 42 CFR §493.1253(b)(2) (2013).

143. Korf J. Release of endogenous amino acids, dopamine, and cyclic AMP from the rat brain: methodological aspects and mutual interferences. *Ann N Y Acad Sci*. 1986;473:418–433. doi:10.1111/j.1749-6632.1986.tb23633.x

144. Gulley JL, Rajan A, Spigel DR, et al. Avelumab for patients with previously treated metastatic or recurrent non-small-cell lung cancer (JAVELIN Solid Tumor): dose-expansion cohort of a multicentre, open-label, phase 1b trial. *Lancet Oncol*. 2017;18(5):599–610. doi:10.1016/s1470-2045(17)30240-1

145. Song L, Zeng L, Yan H, et al. Validation of E1L3N antibody for PD-L1 detection and prediction of pembrolizumab response in non-small-cell lung cancer. *Commun Med (Lond)*. 2022;2(1):137. doi:10.1038/s43856-022-00206-4

146. Gettinger SN, Horn L, Gandhi L, et al. Overall survival and long-term safety of nivolumab (anti-programmed death 1 antibody, BMS-936558, ONO-4538) in patients with previously treated advanced non-small-cell lung cancer. *J Clin Oncol*. 2015;33(18):2004–2012. doi:10.1200/jco.2014.58.3708

147. Carbognin L, Pilotto S, Milella M, et al. Differential activity of nivolumab, pembrolizumab and MPDL3280A according to the tumor expression of programmed death-ligand-1 (PD-L1): sensitivity analysis of trials in melanoma, lung and genitourinary cancers. *PLoS One*. 2015;10(6):e0130142. doi:10.1371/journal.pone.0130142

148. Sezer A, Kilickap S, Gümüş M, et al. Cemiplimab monotherapy for first--line treatment of advanced non-small-cell lung cancer with PD-L1 of at least 50%: a multicentre, open-label, global, phase 3, randomised, controlled trial. *Lancet*. 2021;397(10274):592–604. doi:10.1016/s0140-6736(21)00228-2

149. Singal G, Miller PG, Agarwala V, et al. Association of patient characteristics and tumor genomics with clinical outcomes among patients with non-small cell lung cancer using a clinicogenomic database. *JAMA*. 2019;321(14):1391–1399. doi:10.1001/jama.2019.3241

150. Aggarwal C, Thompson JC, Chien AL, et al. Baseline plasma tumor mutation burden predicts response to pembrolizumab-based therapy in patients with metastatic non-small cell lung cancer. *Clin Cancer Res*. 2020;26(10):2354–2361. doi:10.1158/1078-0432.CCR-19-3663

151. Liu L, Bai X, Wang J, et al. Combination of TMB and CNA stratifies prognostic and predictive responses to immunotherapy across metastatic cancer. *Clin Cancer Res*. 2019;25(24):7413–7423. doi:10.1158/1078-0432.CCR-19-0558

152. Chang H, Sasson A, Srinivasan S, et al. Bioinformatic methods and bridging of assay results for reliable tumor mutational burden assessment in non-small-cell lung cancer. *Mol Diagn Ther*. 2019;23(4):507–520. doi:10.1007/s40291-019-00408-y

153. Suzuki S, Haratani K, Hayashi H, et al. Association of tumour burden with the efficacy of programmed cell death-1/programmed cell death ligand-1 inhibitors for treatment-naive advanced non-small-cell lung cancer. *Eur J Cancer*. 2022;161:44–54. doi:10.1016/j.ejca.2021.11.011

154. Hellmann MD, Ciuleanu TE, Pluzanski A, et al. Nivolumab plus ipilimumab in lung cancer with a high tumor mutational burden. *N Engl J Med*. 2018;378(22):2093–2104. doi:10.1056/NEJMoa1801946

155. Marabelle A, Le DT, Ascierto PA, et al. Efficacy of pembrolizumab in patients with noncolorectal high microsatellite instability/mismatch repair-deficient cancer: results from the phase II KEYNOTE-158 study. *J Clin Oncol*. 2020;38(1):1–10. doi:10.1200/jco.19.02105

156. Keytruda [prescribing information]. Merck Sharp & Dohme BV; 2020.

157. Lamberti G, Spurr LF, Li Y, et al. Clinicopathological and genomic correlates of programmed cell death ligand 1 (PD-L1) expression in nonsquamous non-small-cell lung cancer. *Ann Oncol*. 2020;31(6):807–814. doi:10.1016/j.annonc.2020.02.017

158. Hsu ML, Murray JC, Psoter KJ, et al. Clinical features, survival, and burden of toxicities in survivors more than one year after lung cancer immunotherapy. *Oncologist*. 2022;27(11):971–981. doi:10.1093/oncolo/oyac140

159. Ruste V, Goldschmidt V, Laparra A, et al. The determinants of very severe immune-related adverse events associated with immune checkpoint inhibitors: a prospective study of the French REISAMIC registry. *Eur J Cancer*. 2021;158:217–224. doi:10.1016/j.ejca.2021.08.048

160. Jhun I, Shepherd D, Hung YP, Madrigal E, Le LP, Mino-Kenudson M. Digital image analysis for estimating stromal CD8+ tumor-infiltrating lymphocytes in lung adenocarcinoma. *J Pathol Inform*. 2021;12:28. doi:10.4103/jpi.jpi_36_20

161. Berry S, Giraldo NA, Green BF, et al. Analysis of multispectral imaging with the AstroPath platform informs efficacy of PD-1 blockade. *Science*. 2021;372(6547):eaba2609. doi:10.1126/science.aba2609

162. Park S, Ock CY, Kim H, et al. Artificial intelligence-powered spatial analysis of tumor-infiltrating lymphocytes as complementary biomarker for immune checkpoint inhibition in non-small-cell lung cancer. *J Clin Oncol*. 2022;40(17):1916–1928. doi:10.1200/jco.21.02010

163. Larijani B, Miles J, Ward SG, Parker PJ. Quantification of biomarker functionality predicts patient outcomes. *Br J Cancer*. 2021;124(10):1618–1620. doi:10.1038/s41416-021-01291-3

164. Ghaffari Laleh N, Ligero M, Perez-Lopez R, Kather JN. Facts and hopes on the use of artificial intelligence for predictive immunotherapy biomarkers in cancer. *Clin Cancer Res*. 2023;29(2):316–323. doi:10.1158/1078-0432.Ccr-22-0390

165. Santaguida S, Richardson A, Iyer DR, et al. Chromosome mis-segregation generates cell-cycle-arrested cells with complex karyotypes that are eliminated by the immune system. *Dev Cell*. 2017;41(6):638–651.e5. doi:10.1016/j.devcel.2017.05.022

166. Otano I, Ucero AC, Zugazagoitia J, Paz-Ares L. At the crossroads of immunotherapy for oncogene-addicted subsets of NSCLC. *Nat Rev Clin Oncol*. 2023;20(3):143–159. doi:10.1038/s41571-022-00718-x

167. Baxi V, Lee G, Duan C, et al. Association of artificial intelligence-powered and manual quantification of programmed death-ligand 1 (PD-L1) expression with outcomes in patients treated with nivolumab +/− ipilimumab. *Mod Pathol*. 2022;35(11):1529–1539. doi:10.1038/s41379-022-01119-2

168. Dercle L, McGale J, Sun S, et al. Artificial intelligence and radiomics: fundamentals, applications, and challenges in immunotherapy. *J Immunother Cancer*. 2022;10(9):e005292. doi:10.1136/jitc-2022-005292

169. Wu N, Ge W, Quek RG, et al. Trends in real-world biomarker testing and overall survival in US patients with advanced non-small-cell lung cancer. *Future Oncol*. 2022;18(39):4385–4397. doi:10.2217/fon-2022-0540

170. Gadgeel S, Rodriguez-Abreu D, Speranza G, et al. Updated analysis from KEYNOTE-189: pembrolizumab or placebo plus pemetrexed and platinum for previously untreated metastatic nonsquamous non-small-cell lung cancer. *J Clin Oncol*. 2020;38(14):1505–1517. doi:10.1200/JCO.19.03136

171. Paz-Ares L, Vicente D, Tafreshi A, et al. A randomized, placebo-controlled trial of pembrolizumab plus chemotherapy in patients with metastatic squamous NSCLC: protocol-specified final analysis of KEYNOTE-407. *J Thorac Oncol*. 2020;15(10):1657–1669. doi:10.1016/j.jtho.2020.06.015

172. Carbone DP, Reck M, Paz-Ares L, et al. First-line nivolumab in stage IV or recurrent non-small-cell lung cancer. *N Engl J Med*. 2017;376(25):2415–2426. doi:10.1056/NEJMoa1613493

173. Socinski MA, Nishio M, Jotte RM, et al. IMpower150 final overall survival analyses for atezolizumab plus bevacizumab and chemotherapy in first-line metastatic nonsquamous NSCLC. *J Thorac Oncol*. 2021;16(11):1909–1924. doi:10.1016/j.jtho.2021.07.009

174. Jotte R, Cappuzzo F, Vynnychenko I, et al. Atezolizumab in combination with carboplatin and nab-paclitaxel in advanced squamous NSCLC (IMpower131): results from a randomized phase III trial. *J Thorac Oncol*. 2020;15(8):1351–1360. doi:10.1016/j.jtho.2020.03.028

175. Nishio M, Barlesi F, West H, et al. Atezolizumab plus chemotherapy for first-line treatment of nonsquamous NSCLC: results from the randomized phase 3 IMpower132 trial. *J Thorac Oncol*. 2021;16(4):653–664. doi:10.1016/j.jtho.2020.11.025

APPENDIX

DISCLOSED INTERESTS AND ACTIVITIES FROM JULY 2018 TO OCTOBER 2023

Mark Awad, MD, PhD

Consulting fees or Advisory Board: Affini-T Therapeutics Inc, AstraZeneca, Bristol-Myers Squibb, EMD Serono Inc, Genentech, Gritstone bio, Foundation Medicine, Instil Bio, Janssen Oncology, Merck, Mirati Therapeutics Inc, Novartis, Pfizer, Regeneron Pharmaceuticals

Research grants: AstraZeneca, Bristol-Myers Squibb

David M. Hwang, MD, PhD

Consulting fees or Advisory Board: Amgen Inc, Bayer Canada, GSK Canada, Merck Canada Inc, Novartis Canada, Roche Canada

Research grants: Boehringer Ingelheim

Speakers' bureau/lecture fees/honoraria: Amgen Canada Inc, AstraZeneca Canada, Merck Canada Inc, Pfizer Canada Inc

Gregory Kalemkerian, MD

Research grants: Blueprint Medicines, Cullinan Oncology, Daiichi Sankyo, Merck, Takeda Pharmaceuticals

Fernando Lopez-Rios, MD, PhD

Consulting fees or Advisory Board: AstraZeneca, Bayer, Bristol-Myers Squibb, Daiichi Sankyo, Janssen Oncology, Eli Lilly, Merck Sharp & Dohme Corp, Roche

Research grants: Eli Lilly, Pfizer, Roche Thermo Fisher Scientific

Speakers' bureau/lecture fees/honoraria: AstraZeneca, Bayer, Bristol-Myers Squibb, Eli Lilly, Janssen Oncology, Merck Sharp & Dohme Corp, Pfizer, Roche, Takeda Pharmaceuticals, Thermo Fisher Scientific

Mari Mino-Kenudson, MD

Consulting fees or Advisory Board: AstraZeneca, Janssen Oncology, Sanofi

Lauren Ritterhouse, MD, PhD

Employment: Foundation Medicine*

Speakers' bureau/lecture fees/honoraria: Astellas Pharma Inc, EMD Serono Inc, Sanofi, Thermo Fisher Scientific

Stock options/bonds: Foundation Medicine*

*Employment and stock options were obtained after completion of the guideline manuscript.

Lynette M. Sholl, MD

Consulting fees or Advisory Board: AstraZeneca, Eli Lilly, Genentech

Research grants: Bristol-Myers Squibb, Genentech

Paul E. Swanson, MD

Consulting fees or Advisory Board: Wolters-Kluwer Publishing

No Disclosures to Report

Upal Basu Roy, PhD, MS, MPH, Mary Beth Beasley, MD, Richard Walter Cartun, PhD, MS, Larissa V. Furtado, MD, Ajit Paintal, MD, Kearin Reid, MLS(ASCP), MLIS, Lesley A. Souter, PhD, Christina B. Ventura, MPH, MT(ASCP)