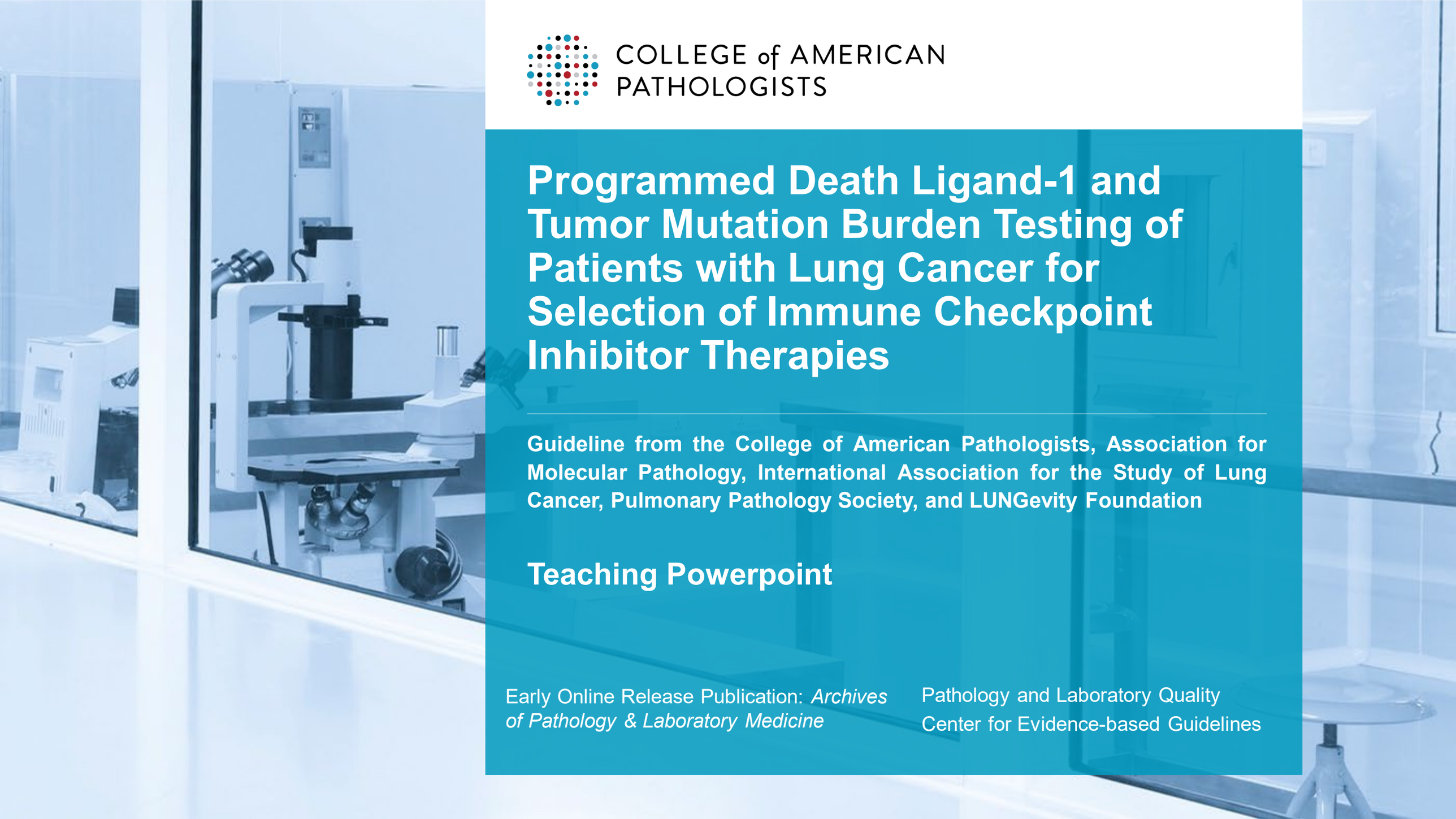


COLLEGE of AMERICAN PATHOLOGISTS

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

Teaching Powerpoint

Early Online Release Publication: *Archives of Pathology & Laboratory Medicine*

Pathology and Laboratory Quality
Center for Evidence-based Guidelines

Outline

- Introduction
- Objectives
- Key Questions and Results
- Guideline Recommendations
- Guideline Development Process
- Conclusions

Introduction

Introduction

- The clinical impact of immune checkpoint inhibitors (ICI) are profound for patients with non-small cell lung carcinoma (NSCLC).
- Clinical trials 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.
- Testing these PD-L1 antibodies are particularly complicated with the individual therapies, along with different tumor types, and companion diagnostics approved by agencies such as the US FDA.

Introduction, continued

- Due to the methodologic variabilities of different testing modalities, there can be some uncertainty in the test selection and implementation of PD-L1 testing.
- The College of American Pathologists convened a panel of experts in NSCLC 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.

Objectives

Objectives

- 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

Key Questions and Results

Key Questions

- 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?
- When selecting patients for anti-PD1 and anti-PD-L1 therapy, does testing of different specimen types provide concordant clinical outcomes?
- Does the use of ICI therapy in patients with advanced NSCLC with targetable *ALK*, *EGFR*, *ROS1*, or *BRAF* molecular alterations affect their long-term clinical outcomes?

Key Questions, continued

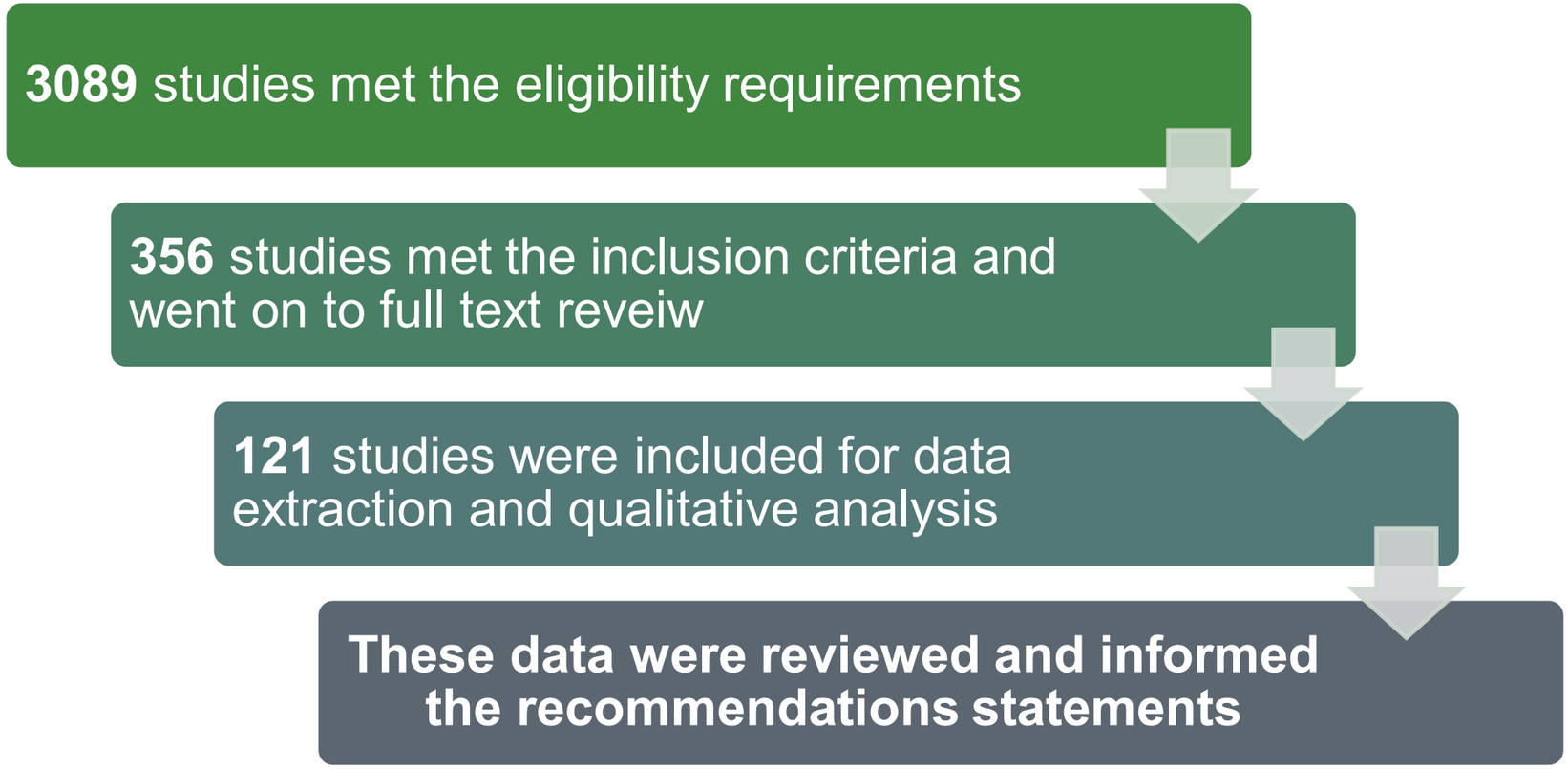
- When selecting patients for anti-PD-1 and anti-PD-L1 therapy, does TMB testing have the analytical validity to identify a complementary population who will benefit from therapy?
- In patients with NSCLC with more than one available sample, do multiple samples provide concordant PD-L1 and TMB testing results and downstream clinical outcomes?
- Does clinical validity of PD-L1 testing differ by levels of PD-L1 expression in tumor or immune cells?

Key Questions, continued

- How reproducible are PD-L1 tumor cell scores and immune cell scores across specimen types?
- Do the available PD-L1 assays provide concordant expression profiles when evaluating the same sample and which IHC expression cut-off provides the most reproducible expression categorization across the assays?

Results

- **To address the Key Questions, a systematic literature review was performed. Six recommendations were drafted.**



Guideline Recommendations

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 immune checkpoint inhibitors. *(Strength of Recommendation: Strong; Certainty of Evidence: Moderate).*

Rationale: Data show significant correlation between the 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

Overall Survival and Response Rates for First Line Immune Checkpoint Inhibitors

RCT	FDA Approval (Y / N)	Treatment Arms	PD-L1 IHC Clone	Overall Response Rate	Overall Survival
KEYNOTE-024, Reck et al, 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, 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, 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, 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, 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, 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, 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

Overall Survival and Response Rates for First Line Immune Checkpoint Inhibitors

RCT	FDA Approval (Y / N)	Treatment Arms	PD-L1 IHC Clone	Overall Response Rate	Overall Survival
CheckMate 227, Hellmann et al,2019	Y	Nivolumab + ipilimumab, n=396 Chemotherapy, n=396	28-8	TPS≥1A%: 35.9%; 95%CI, 31.1-40.8%	TPS ≥1%[A]: HR, 0.79; 95%CI, 0.65-0.96; *P*=.007
				TPS ≥50%: 44.4%; 95%CI, 37.5-51.1%	TPS ≥50% HR, 0.70; 95%CI, 0.55-0.90
IMpower110, Herbst et al, 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, 2021	Y	Atezolizumab + Chemotherapy, n=402 Chemotherapy, n=400	SP142	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
		Atezolizumab + Chemotherapy, n=299 Chemotherapy, n=279	SP263	Not reported	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, 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

Overall Survival and Response Rates for First Line Immune Checkpoint Inhibitors

RCT	FDA Approval (Y / N)	Treatment Arms	PD-L1 IHC Clone	Overall Response Rate	Overall Survival
IMpower 132, Nishio et al, 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
IMpower 130, West et al,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

Recommendation 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 for details on how to validate IHC specimens.

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

Rationale: There is variability in the different types of specimen that can be tested for PD-L1 (specimen age, site, size, modality of preparation); the panel recommends that validation should be done on all the different types of specimen and fixatives according to the requirements of the laboratory's accreditation agency.

Recommendation 3

When feasible, pathologists should use clinically validated PD-L1 IHC assays as intended. *(Strength of Recommendation: Conditional: Certainty of Evidence: Very low)*

Rationale: The concordance of PD-L1 expression is 90% or greater for 22C3, 288, and SP263 companion diagnostic assays (CDX), while VENTANA SP142 shows weaker expression.

Interobserver agreement/reproducibility on tumor cells scoring is similar among all 4 mentioned CDx assays. The EP recommends validation according to the manufacturer recommendations to ensure clinical performance of these assays.

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	TC ≥1%	Atezolizumab

PD-L1 Status Concordance in FDA-Approved Companion Diagnostic Assays and Laboratory Developed Tests

FDA Approved CDx[A]	LDT (clone; platform)	TPS Cut-off		Concordance (range reported by included studies)
		CDx	LDT	
22C3 PharmDx	73-10; Dako/Agilent	≥50%	≥80%	93.9% Grote et al, 2020
	E1L3N; VENTANA	≥50%	≥50%	88.2% Munari et al, 2019
SP263 Assay	22C3; VENTANA	≥1%	≥1%	89.7 – 100% Munari et al, 2018, Sughayer et al, 2019
	E1L3N; VENTANA	≥50%	≥50%	95.8% Munari et al, 2019

A. FDA approval criteria are outlined in previous table.

Kappas for PD-L1 Status in FDA-Approved Companion Diagnostic Assays and Laboratory Developed Tests

FDA Approved CDx[A]	LDT (clone; platform)	TPS Cut-off		Kappa (range reported by included studies)
		CDx	LDT	
SP263 Assay	22C3; VENTANA	≥50%	≥50%	0.73 – 0.75, Munari et al, 2018
	SP263; Dako/Agilent	≥1%	≥1%	0.83 – 0.86, Adam et al, 2018
	SP263; Leica	≥1%	≥1%	0.83 – 0.86, Adam et al, 2018
	SP142; Dako/Agilent	≥1%	≥1%	0.38 – 0.68, Adam et al, 2018
	SP142; Leica	≥1%	≥1%	0.78 – 0.81, Adam et al, 2018
	E1L3N; Dako/Agilent	≥1%	≥1%	0.63 – 0.77, Adam et al, 2018
	E1L3N; VENTANA	≥1%	≥1%	0.60 – 0.81, Adam et al, 2018
	E1L3N; Leica	≥1%	≥1%	0.75 – 0.78, Adam et al, 2018
SP142 Assay	E1L3N; VENTANA	≥1%	≥1%	0.65, Kim et al, 2017
22C3 Assay	22C3; VENTANA	≥1%	≥1%	0.77 – 0.81, Adam et al, 2018
	22C3; Leica	≥1%	≥1%	0.50 – 0.62, Adam et al, 2018
28-8 Assay	28-8; VENTANA	≥1%	≥1%	0.73 – 0.80, Adam et al, 2018
	28-8; Leica	≥1%	≥1%	0.58 – 0.60, Adam et al, 2018

Recommendation 4

Pathologists that choose to use laboratory developed tests (LDTs) for PD-L1 expression should validate according to the requirements of their accrediting body. *(Strength of Recommendation: Strong; Certainty of Evidence: Very Low)*

Rationale: A LDT is defined as an in vitro diagnostic test that is designed and used within a single laboratory. A LDT should be accurate to ensure that patients get appropriate and timely treatments, thus assay validation is recommended.

LDTs using FDA Approved Antibodies

Approved Assay	Modified PD-L1 Expression Cut Point	Modified Platform
22C3 assay	TPS 1-49%, Gadgeel et al, 2020 Herbst et al, 2021	VENTANA, Munari et al, 2018, Villaruz et al, 2019, Sughayer et al, 2019, Ilie et al, 2018, Adam et al, 2018
	TPS ≥50%, Reck et al, 2021, Mok et al, 2019, Gadgeel et al, 2020, Herbst et al, 2021	Leica, Adam et al, 2018
28-8 assay	TPS ≥5%, Carbone et al, 2017	VENTANA, Adam et al, 2018
	TPS ≥50%, Brahmer et al, 2015, Hellmann et al, 2019	Leica, Adam et al, 2018
SP142 assay	TC1/2 or IC1/2, West et al, 2019	Dako/Agilent, Adam et al, 2018
	TC2/3 or IC2/3, Herbst et al, 2020	Leica, Adam et al, 2018
	TC1-3 or IC1-3, Socinski et al, 2021	
SP263 assay	None reported	Dako/Agilent, Adam et al, 2018
		Leica, Adam et al, 2018

LDTs 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, 2018, Grote et al, 2020, Park et al, 2021	Avelumab, Barlesi et al, 2018, Grote et al, 2020, Park et al, 2021
		TPS ≥50%, Barlesi et al, 2018, Grote et al, 2020, Park et al, 2021	
		TPS ≥80%, Barlesi et al, 2018, Grote et al, 2020, Park et al, 2021	
E1L3N	VENTANA	TPS ≥1%, Kim et al, 2017, Munari et al, 2019	Atezolizumab, Kim et al, 2017, Munari et al, 2019
		TPS ≥5%, Kim et al, 2017	
		TPS ≥50%, Kim et al, 2017, Munari et al, 2019	

Recommendation 5

Pathologists should report PD-L1 immunohistochemistry results using a percent expression score. *(Strength of Recommendation: Conditional; Certainty of Evidence: Very low)*

Rationale: Providing an exact percent expression score value may be challenging due to the subjective nature of visual assessment of PD-L1 expression and scoring variability among pathologists. One option that several clinical laboratories have adopted is to report ranges of PD-L1 percent expression scores (eg, 5% or 10% incremental values) instead of absolute scores. A semi-quantitative approach can more accurate and reproducible than reporting specific expression percent values while providing information for management decisions.

Recommendation 6

Clinicians should not use TMB 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)*

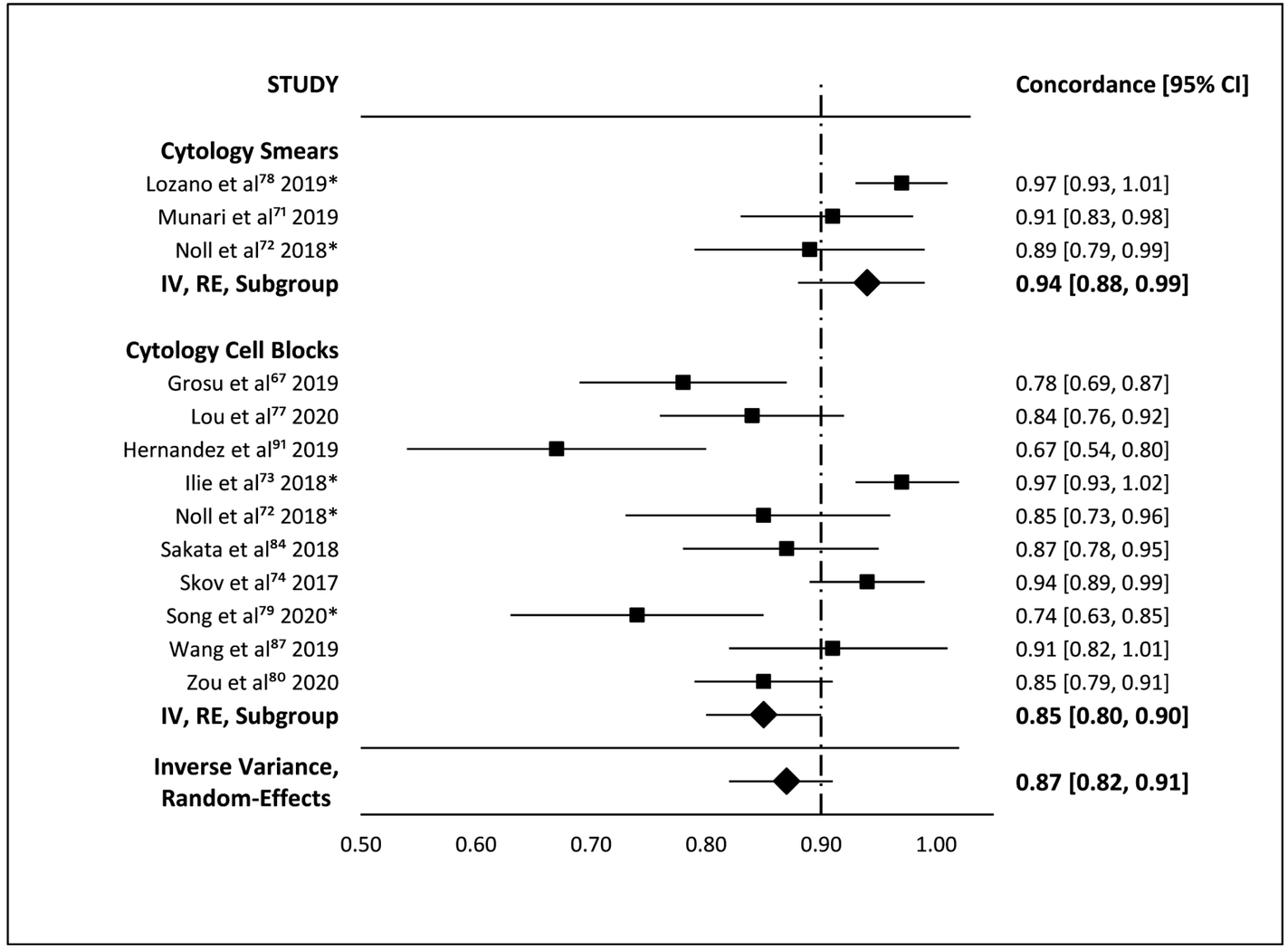
Rationale: The CheckMate-22716 trial showed an improved 1-year PFS rate for patients with advanced NSCLC and TMB ≥10 mutations/megabase (mut/Mb) receiving nivolumab plus ipilimumab in the first line setting; the supplemental FDA application was withdrawn when subsequent data showed no difference in survival outcomes between patients stratified by high or low tumor TMB. The FDA approval for nivolumab plus ipilimumab as first line treatment for patients with metastatic NSCLC requires tumor PD-L1 expression of ≥1%, as determined by an FDA-approved test, with no EGFR and ALK genomic tumor aberrations, and **does not include TMB**.

Recommendation 6, continued

Clinicians should not use TMB 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)*

Rationale: Accelerated approval was granted to pembrolizumab for the treatment of adult and pediatric patients with unresectable or metastatic TMB-high (≥10 mut/Mb) solid tumors that have progressed following prior treatment and **who have no satisfactory alternative treatment options.** The KEYNOTE-158 study included 102 patients with TMB ≥10 mut/Mb spanning 9 different tumor types, none of which were NSCLC.

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 biopsies, and surgical sections.

Abbreviations: CI, confidence interval; IV, inverse variance; PD-L1, programmed death ligand-1; RE, random effects; FFPE, formalin-fixed, paraffin-embedded.

Guideline Development Process

Expert Panel (EP)

- Lynette M. Sholl, MD*
- Larissa V. Furtado, MD*
- Mark Awad, MD, PhD
- Mary Beth Beasley, MD (PPS)
- Richard Walter Cartun, PhD, MS
- David M. Hwang, MD, PhD
- Gregory Kalemkerian, MD (ASCO)
- Fernando Lopez-Rios, MD, PhD (IASLC)
- Mari Mino-Kenudson, MD
- Ajit Paintal, MD
- Lauren Ritterhouse, MD, PhD (AMP)
- Lesley A. Souter, PhD
- Paul E. Swanson, MD

CAP Staff
Christina B. Ventura, MPH, MT(ASCP)
Senior Guideline Development Manager
Kearin Reid, MLS(ASCP), MLIS
Medical Librarian Specialist

Ms. Reid was employed by the CAP during the development process.

*Guideline Chair
AMP, Association for Molecular Pathology; ASCO, American Society for Clinical Oncology; IASLC, International Association for the Study of Lung Cancer, PPS, Pulmonary Pathology Society

Advisory Panel (AP)

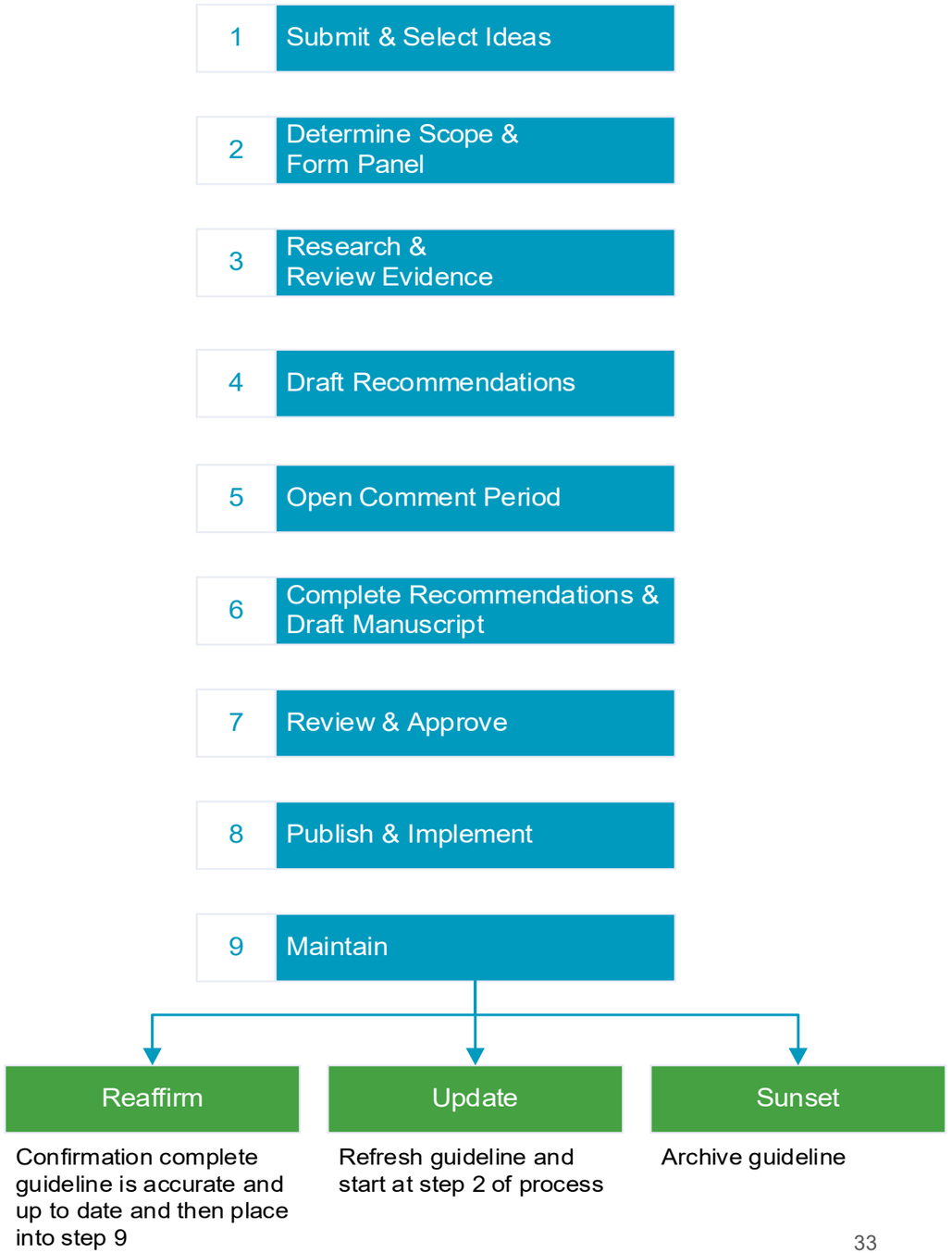
- Ezra Baraban, MD
- Eric Bernicker, MD
- Russell Broaddus, MD, PhD
- Sanja Dacic, MD, PhD
- Fang Fan, MD, PhD
- Patrick Fitzgibbons, MD
- Zaibo Li, MD, PhD
- Robert McGee, MD
- Sinchita Roy-Chowdhuri, MD, PhD
- Marina Vivero, MD
- Ahmet Zehir, PhD

LUNGevity Foundation Patient Advocates

- Upal Basu Roy, PhD, MS, MPH
- Barbara A. Ward

Guideline Development Process

- The Center follows the standards endorsed by the National Academy of Medicine for developing Clinical Practice Guidelines.
- Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach was utilized in updating the guideline.
- A detailed description of the guideline development process can be found online [Evidence-based Guidelines Development Methodology Manual](#) .



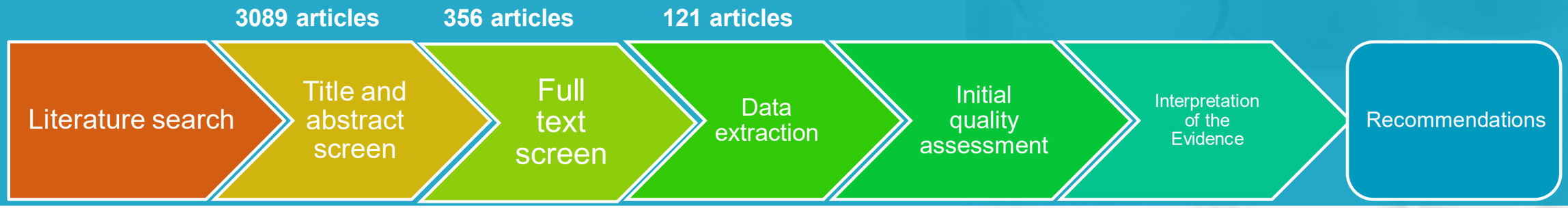
Literature Search and Systematic Review

Search was conducted in Ovid MEDLINE, Embase, Cochrane Library.

- **Literature search ran on: 10/16/2019 and rerun on 4/7/2021 and 5/13/2022**
- **A total of 3089 studies were captured**

Literature Search and Systematic Review

- Search was conducted in Ovid MEDLINE, Embase, Cochrane Library.
- Literature search ran on: 10/16/2019 and rerun on 4/7/2021 and 5/13/2022
- A total of 3089 studies were captured
- Each level of systematic review (title-abstract screening, full-text review, and data extraction) was performed in duplicate by two members of the expert panel.



Methods

- This evidence-based guideline was developed following the standards by the National Academy of Medicine
- The CAP collaborated with AMP, ASCO, IASLC, PPS and LUNGevity Foundation and convened a multidisciplinary expert and advisory panel to develop the guideline
- The overarching questions, “Does PD-1/PD-L1 status and tumor mutation burden 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?” was addressed by the EP

Panel Proceedings

- The expert panel met via conference call/webinar multiple times and once in-person throughout the development of the guideline to develop the scope, draft recommendations, review and respond to solicited feedback, and assess the certainty of evidence that supports the final recommendations

Panel Proceedings, continued

- Open Comment Period held from March 31 to April 23, 2021
- Six draft statements, demographic questions, and questions to assess feasibility were posted for peer review
- Over 130 respondents participated, and the guideline received 228 individual comments
- All 6 recommendations received between 91.67 – 95.65% Agree or Agree with suggested modifications

Panel Proceedings, continued

- An independent review panel (IRP) was assembled to review and approve the guideline on behalf of the CAP Council on Scientific Affairs.
- The IRP was masked to the EP and to each other and were vetted through the COI process.
- Collaborating organizations were provided the guideline for approval.

Conclusions

Conclusions

- PD-L1 IHC testing is 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.
- Regulatory-approved, clinically validated PD-L1 diagnostics are recommended. However, the panel also recognized that the practical reality of most laboratories may require use of LDTs. Therefore, the panel also endorses the use of LDTs provided that the assays are adequately validated.

Conclusions

- Laboratories need to recognize the different variables that influence PD-L1 expression status (eg, expression heterogeneity, sample requirements, and appropriate validation for each of the sample types) to render an accurate diagnosis for PD-L1.
- 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 published data is insufficient to date to suggest that the current cut point for TMB-high is a reliable predictor of ICI response.

References

Sholl LM, Furtado LV, Awad M, et al. 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, the Association for Molecular Pathology, the International Association for the Study of Lung Cancer, the Pulmonary Pathology Society, and the LUNGevity Foundation. *Arch Pathol Lab Med.* Published online April 16, 2024.

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- 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, express 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.

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