# Immunotherapy in Lung Cancer Treatment: A New Paradigm Still in Revision

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***Purpose and Overview:*** To discuss changing paradigms in the treatment of advanced lung cancer with immune checkpoint blockade. Antibodies against inhibitory T cell immune checkpoints have now become standard of treatment in the first- and second-line therapies for metastatic lung cancer. We will review the role of T cell immune check points in tumor biology, a brief overview of the clinical data of immune checkpoint inhibitors, attempts to identify predictive biomarkers, and a discussion of immune related adverse events from T cell checkpoint blockade.

***Objectives:***

1. To review biology of T cell immune checkpoint blockade

2. To review the new immune checkpoint inhibitors and their roles in the treatment of lung cancers.

3. To review controversies in identification of predictive biomarkers for the immune checkpoint inhibitors.

4. To review the immune-related adverse events from immune checkpoint blockade and their management.

## INTRODUCTION

Lung cancer is the most common cause of cancer death worldwide for both men and women. In the U.S., it is estimated that there will be nearly as many deaths due to lung cancer than breast, prostate, colon and pancreas cancers combined in 2013 [1]. This is despite lung cancer being the second most common cancer diagnosis behind breast and prostate cancer in women and men, respectively [2]. Less than 17% of all lung cancer patients are alive five years after their diagnosis [3]. For patients with localized disease, five year survival rates are still poor (54%) and much worse (< 4%) for patients with metastatic disease [3].

The major groups of lung cancer traditionally have been divided into small cell and non-small cell lung cancer (NSCLC). Small cell lung cancer is now considered to be the most aggressive form of a larger group of pulmonary neuroendocrine tumors. Tumors in this spectrum also include typical and atypical pulmonary carcinoid tumors and large cell neuroendocrine tumors. Non-small cell lung cancer consists of three major histologic subtypes: adenocarcinoma, squamous cell carcinoma, and large cell carcinoma (without neuroendocrine features).

Adenocarcinoma is now the most common subtype accounting for ~40% of lung cancers. Adenocarcinomas are still most common among current and former smokers. However, among non-smokers who develop NSCLC, the tumors are disproportionately adenocarcinomas[4]. Adenocarcinomas are often found in the periphery of the lungs, are aggressive, and frequently metastasize. Diagnosis of adenocarcinoma is confirmed through immunohistochemical (IHC) stains. Thyroid transcription factor 1 (TTF-1, NKX 2.1) and napsin A are most commonly used to identify adenocarcinoma. TTF-1 staining is only found in thyroid cancers and ~80% of lung adenocarcinomas.

Squamous cell carcinoma (SCC) is the second most common histologic subtype of NSCLC and is highly correlated with smoking history. The incidence of squamous cell lung cancer has been decreasing in the U.S. due to decreased smoking rates among the population. These tumors are often found centrally near the midline and are the ones most likely to cavitate among the various NSCLC histologic subtypes. IHC stains include the cytokeratins CK 5/6 and the transcription factor p63. These tumors are often negative for TTF-1.

Large cell carcinomas make up <10% of NSCLC. Their histologic appearance consists of sheets of undifferentiated cells without glandular or squamous differentiation with prominent nucleoli. Necrosis is often seen with these aggressive tumors. The incidence of large cell NSCLC has decreased substantially as IHC has improved to distinguish adenocarcinomas, squamous cell carcinomas and pulmonary neuroendocrine cancers.

Small cell lung cancer (SCLC) comprises ~13% of lung cancer and is strongly correlated with smoking. Similar to SCC, the incidence of SCLC has been decreasing as smoking rates have decreased. SCLC is the most aggressive form of pulmonary neuroendocrine cancers with metastases identified in the majority of cases at diagnosis. IHC stains include TTF-1 and the neuroendocrine markers chromogranin, synaptophysin and CD56 (NCAM).

## HISTORICAL HIGHLIGHTS OF THERAPY FOR METASTATIC NSCLC

**Chemotherapy Era.** We will focus here on therapy for metastatic NSCLC. Prior to the 1990s, the use of chemotherapy for NSCLC was controversial. Numerous chemotherapy trials

during the 1970s and 1980s failed to show a survival benefit over best-supportive care due to lack of effective treatments, intolerable side effects, and the inadequate statistical power of these studies to detect modest benefits. With best-supportive care at that time, median survival was 4-5 months and 1 year-survival was 10%. In 1995, a landmark meta-analysis analyzed 11 trials with 1190 patients comparing chemotherapy against supportive care in the metastatic setting [5]. The analysis established the benefit of cisplatin-based regimens with increase in median survival by 1.5 months and a hazard ratio for death of 0.73 ($p<0.001$) in favor of cisplatin-based chemotherapy.

Another landmark study [6] in 2002 compared the four regimens of cisplatin/gemcitabine, cisplatin/docetaxel, carboplatin/paclitaxel, and cisplatin/paclitaxel in 1155 patients with metastatic disease divided equally in each treatment arm and found no significant difference in overall survival and time to disease progression (Fig. 1). These four regimens became the standard treatments for metastatic NSCLC regardless of histologic subtype. The choice of regimen depended upon the toxicity profile that was most compatible with the patient's condition.

Histology emerged as predictive factor with a phase III NSCLC trial that compared cisplatin-gemcitabine (CG) or cisplatin-pemetrexed (CP) treatment arms [7]. Median overall survival (10.3 months for both arms) and progression-free survival (CG 5.1 months, CP 4.8 months) were equivalent for the 2 regimens. However, treatment of non-squamous cell NSCLC (adenocarcinoma and large cell carcinoma) with pemetrexed showed statistically significant improvement in median overall survival and a statistically significant hazard ratio that favored the pemetrexed arm. Conversely, patients with squamous cell histology had a statistically significant improvement in median overall survival with cisplatin-gemcitabine compared with cisplatin-pemetrexed.

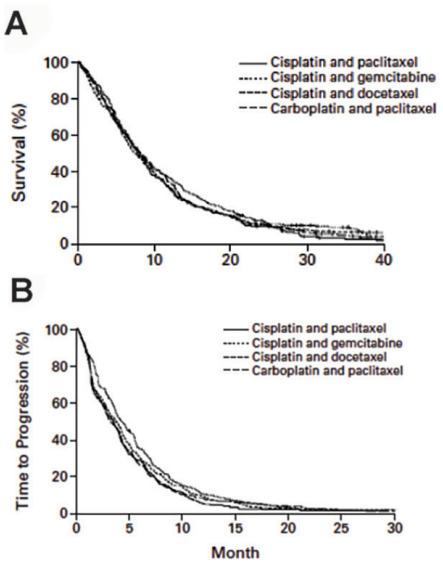



**Figure 1. Equivalence of platinum doublets in NSCLC.** Efficacy of platinum-doublet chemotherapies are equivalent for (A) overall survival and (B) time to progression for NSCLC. *From Schiller et al., 2002.*

**Molecular Targeted Era.** A phase II trial showed efficacy of angiogenesis inhibition by adding an anti-VEGFA antibody, bevacizumab (Avastin; Genentech), to chemotherapy [8]. The results were confirmed in the phase III ECOG 4599 randomized phase III trial [9] using carboplatin-paclitaxel with or without bevacizumab in patients with stage IIIB or IV non-squamous NSCLC. Patients with squamous cell histology were excluded due to increased rates of life-threatening pulmonary hemorrhages in SCC patients in the previous phase II trial [8]. The phase III ECOG trial showed statistically significant improvements in response rate (35% vs. 15%), progression-free survival (6.2 vs. 4.5 months) and overall survival (12.3 vs. 10.3 months). Based on this trial, bevacizumab, in combination with carboplatin and paclitaxel, was approved by the FDA as first-line treatment for unresectable, locally advanced or metastatic non-squamous cell NSCLC in 2006.

In 2004, the EGFR-activating mutations, L858R and exon 19 deletions, in lung adenocarcinoma were identified [10-12] with sensitivity [13-17] to the reversible ATP-competitive small molecule antagonists, gefitinib [18, 19] and erlotinib [20, 21] (Fig. 2). The most common resis-

tance mechanism against gefitinib and erlotinib is an acquired EGFR-T790M mutation that dramatically increases EGFR's affinity for ATP over the EGFR antagonists [22]. Osimertinib [23, 24], a third-generation EGFR antagonist with activity against EFGR-T790M, is now available as first [25] or second [26] line therapies for lung adenocarcinoma with activating EGFR mutations. Second generation EGFR antagonists include afatinib and dacominitib.

A constitutively active fusion protein of anaplastic lymphoma kinase (ALK) fusion protein product is present, mostly with the echinoderm microtubule associated protein like 4 (EML4)[27, 28] are found in ~3-7% of lung adenocarcinomas. Crizotinib, an oral ATP-competitive antagonist of MET with activity against ALK and ROS1 receptor tyrosine kinases, was the first small antagonist to show therapeutic efficacy in ALK-fusion lung adenocarcinoma [29, 30]. Alectinib [31, 32] and brigatinib [33], second generation antagonists with greater affinity for ALK than crizotinib, were shown to be superior to crizotinib in treatment naïve lung adenocarcinoma patients with ALK-fusions. Also, alectinib and brigatinib has demonstrated significant CNS activity that crizotinib lacks. Analogous to EGFR antagonists, resistance to crizotinib develops with acquired ALK resistance mutations or amplifications occurring in ~30% of cases. Alectinib [34], brigatinib [35], and ceritinib [36], another next generation ALK antagonist, have been FDA-approved for patients who have progressed on crizotinib. Lorlatinib has been approved for patients with ALK-fusions who have progressed on crizotinib and a second ALK antagonist. Lorlatinib has activity against all known resistance mutations to ALK antagonists [37, 38].

Constitutively active fusion proteins of ROS1 occur in ~1-2% of lung adenocarcinomas and are susceptible to crizotinib [39] due to the high homology between ROS1 and ALK. Crizotinib and entrectinib [40], a TRK/ROS1/ALK antagonist, have been approved for patients with ROS1-fusion lung adenocarcinoma.

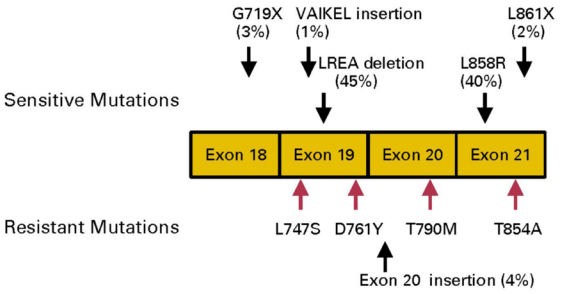



**Figure 2. Schematic of EGFR mutations in tyrosine kinase domain.** Mutations that confer sensitivity to EGFR TKIs are listed above whereas mutations conferring resistance are listed below. Exon 19 deletions and L858R mutations constitute the vast majority of sensitivie mutations. T790M accounts for 90% of the resistance mutations. *From Ohashi et al., JCO 2013.*

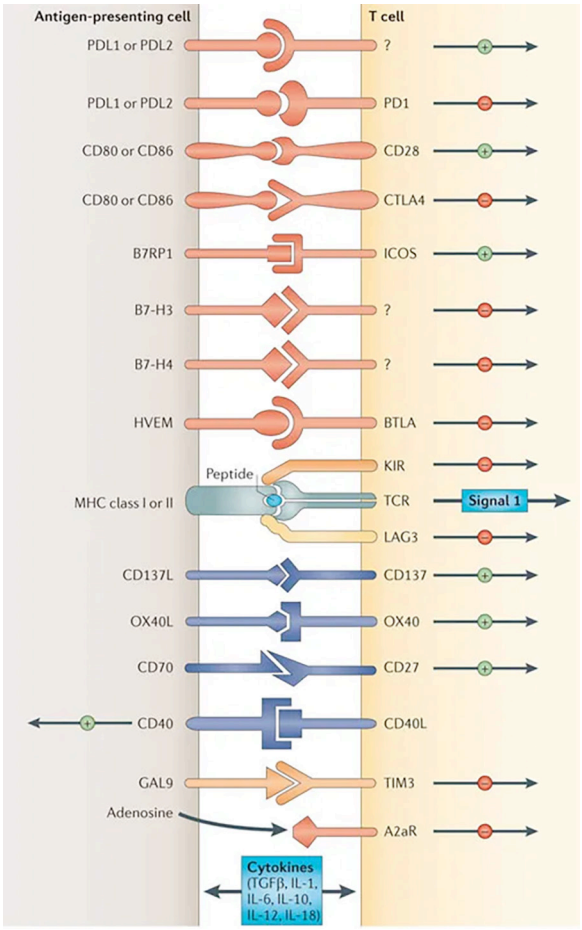



**Figure 3. Schematic of stimulatory and inhibitory T cell checkpoints.** Ligand-receptor interactions that regulate T cell response to antigen stimulation upon binding of the T cell receptor (TCR) to antigen presented on MHC class I or II molecules. '+' are stimulatory signals and '-' are inhibitory signals. *From Pardoll., Nat. Rev. Cancer 2012.*

## ERA OF IMMUNE CHECKPOINT THERAPY

The success of PD-1:PD-L1/2 complex blockade of T cell activation has ushered in a new

therapeutic era for lung cancer, melanoma, renal cell carcinoma, and others. Currently, there are >1,750 active interventional clinical trials across all cancers that involve the PD-1:PD-L1/2 complex and of these, >440 trials are in lung cancer (Clinicaltrials.gov). Currently, two anti-PD-1 (nivolumab, pembrolizumab) and two anti-PD-L1 (atezolizumab, durvalumab) are FDA-approved for treatment of lung cancer patients.

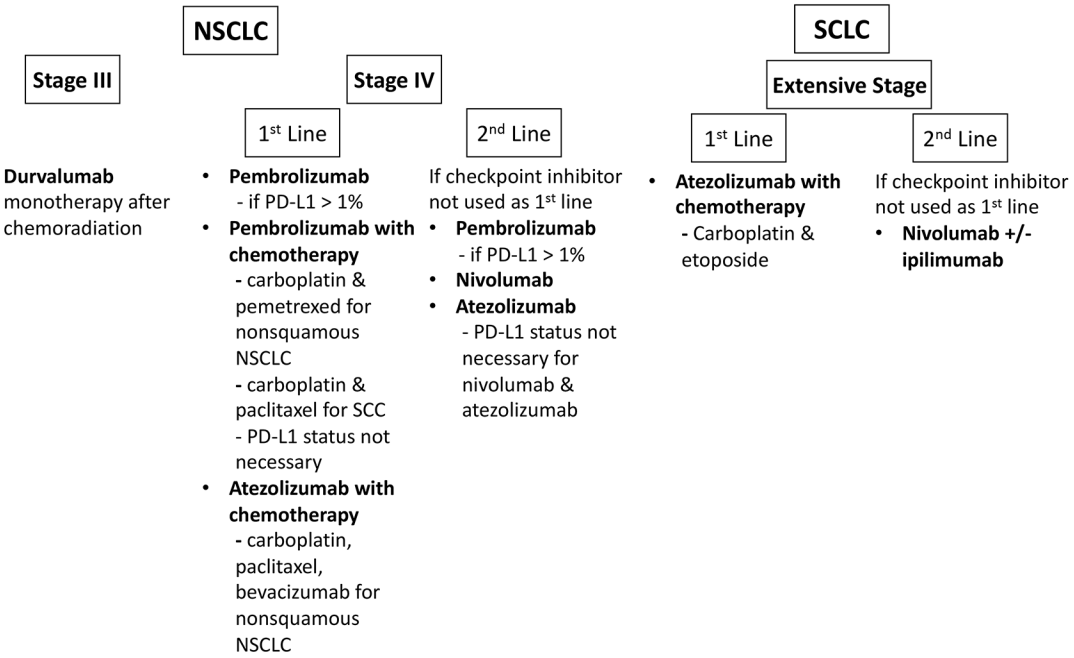
Numerous stimulatory and inhibitory checkpoints for T cell activation have been identified (Fig. 3) [41]. T cell activation requires the binding of the T cell receptor (TCR) to the antigen presented by MHC molecules on antigen presenting cell (APC) and/or tumor cells and the binding of CD28 on T cells with CD80 or CD86 ligands on APCs. CTLA-4 was the first T cell intrinsic inhibitory to be reported [42-45]. James Allison's group demonstrated in vivo tumor regression and immunity to a second exposure of tumors when CTLA-4 was antagonized with antibodies [46].

PD-1 was identified by Tasuku Honjo's group [47] and shown to be an inhibitory receptor [48]. PD-1 is expressed on the cell surface of T cells, B cells, monocytes, dendritic cells, and NK cells. PD-1 binds to PD-L1 and –L2 ligands. PD-L1 is mainly an inducible molecule in humans and found on APCs, macrophages, T and B cells, and tumors. Binding of PD-L1 or –L2 to PD-1 induces PTEN expression to inhibit the PI3K pathway in T cells that leads to decreased cytokine production, proliferation and cell survival and increases the likelihood of immune evasion by the cancer cells.

Allison and Honju shared the 2018 Nobel Prize in Medicine for their work on immune checkpoint therapy for cancer.

**Clinical Trials**

A summary of current indications for immune checkpoint therapy in lung cancer is shown below.




**Phase I Trials.** Two phase I clinical trials with anti-PD-1(BMS-936558, Nivolumab, Bristol Myers

Squibb (BMS)) [49] and anti-PD-L1(BMS-936559, BMS) [50] antibodies showed objective responses of 10-28% in patients with metastatic melanoma, NSCLC, renal-cell carcinoma. Furthermore, some dramatic and long durable responses up to 24 months were noted in patients with melanoma, NSCLC, and renal cell carcinoma - an astounding length of response for a heavily pretreated population (Fig. 4). Long term follow-up of NSCLC patients treated with nivolumab revealed a 5 year overall survival rate of 15.6% [51] (Fig. 5), an increase of nearly four times the survival rate on chemotherapy. Melanoma and renal cell carcinoma were known to be sensitive to immune modulation. However, this was the first demonstration that NSCLC is also responsive to immune therapies. These two trials provided the critical clinical data for the initiation of the immune checkpoint era.

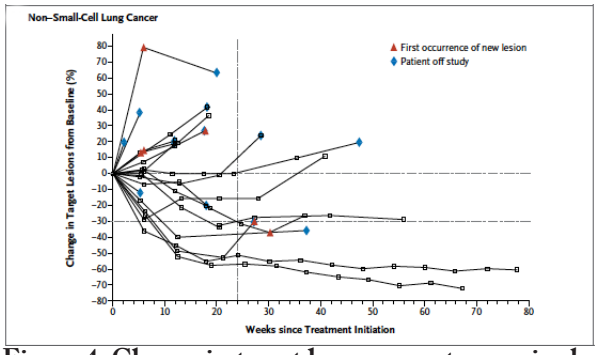



**Figure 4. Change in target lung cancer tumor size by anti-PD-L1 antibody.** Vertical dashed line represent time point when progression free survival was calculated. *From Brahmer et al., NEJM 2012.*

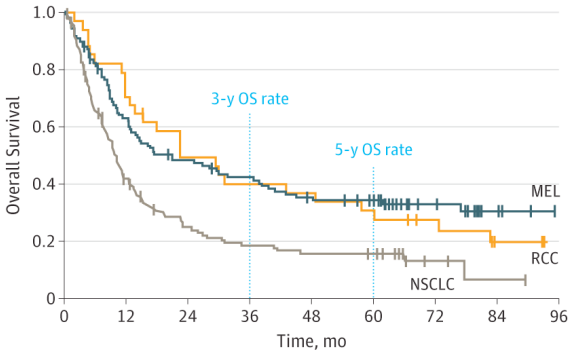



**Figure 5. Long term overall survival on nivolumab.** Overall survival of melanoma (MEL), renal cell carcinoma (RCC) and NSCLC treated with nivolumab in phase I CA209-003 trial. *From Topalian et al., JAMA Oncol. 2019.*

**Second Line Therapy in Advanced Disease.** The first approvals for immune checkpoint therapy in squamous and non-squamous NSCLC were for the anti-PD-1 antibodies, nivolumab (BMS) [52, 53] and pembrolizumab (Merck) [54] in the second line setting after chemotherapy. Both nivolumab and pembrolizumab extended overall survival of NSCLC patients without targetable mutations (i.e. EGFR mutations, ALK- and ROS-1-fusions) over the standard docetaxel chemotherapy. For pembrolizumab, >1% of tumor cells (tumor proportion score (TPS)) are required to be positive for PD-L1 ligand by the DAKO 22C3 PharmaDx immunohistochemistry (IHC) assay. Thereafter, atezolizumab (Genentech/Roche) [55], an anti-PD-L1 antibody, showed superior efficacy compared to docetaxel and was approved in the second line setting.

Second line studies with nivolumab [52, 56] and pembrolizumab [54] have consistently shown worse hazard ratios for overall survival for patients with activating EGFR mutations and ALK-fusions compared to their wild type counterparts. Thus, subsequent immune checkpoint trials excluded patients with these actionable mutations.

In small cell lung cancer (SCLC), nivolumab +/- ipilimumab (anti-CTLA-4 antibody) is approved for patients in the second line setting. Objective response rates were similar between nivolumab alone and in combination with two doses of ipilimumab [57]. Overall survival favored nivolumab with ipilimumab over nivolumab but there was no statistical significance [57].

**First Line Therapy in Advanced Disease.** Pembrolizumab has been FDA-approved for first line therapy alone [58, 59] or in combination with platinum-based chemotherapy [60, 61] for treatment naïve patients with NSCLC. In all studies, pembrolizumab with or without chemotherapy was superior to chemotherapy. Furthermore, response rates correlated with the de-

gree of TPS positivity. PD-L1 TPS > 1% is required for pembrolizumab use as monotherapy. In contrast, pembrolizumab with chemotherapy was superior to chemotherapy for overall survival regardless of TPS score [60, 61]. Atezolizumab in combination with chemotherapy and bevacizumab (anti-VEGF-A antibody) in treatment naïve non-squamous metastatic NSCLC is superior to chemotherapy with bevacizumab [62] and now FDA-approved for that indication, irrespective of PD-L1 status. In contrast, nivolumab as monotherapy was no better than standard chemotherapy in the first line setting for patients with nonsquamous NSCLC [63]. A recent report showed that nivolumab plus ipilimumab (anti-CTLA-4, BMS) was superior to standard chemotherapy in median overall survival (17.1 vs 13.9 months) regardless of PD-L1 status [64].

For extensive stage SCLC, the addition of atezolizumab to standard carboplatin/etoposide chemotherapy improved overall survival [65] although the effect was smaller than those in NSCLC. Atezolizumab in combination with platinum based chemotherapy has been approved by the FDA and is now the new standard first line regimen for extensive stage small cell lung cancer.

**First Line Therapy in Locally Advanced Disease.** Durvalumab (anti-PD-L1 antibody, Medimmune/Astra Zeneca) after concurrent chemotherapy and radiation was superior to concurrent chemotherapy and radiation alone in progression-free survival, objective response rates, and median duration of response for patients with stage III locally advanced NSCLC [66]. For stage III NSCLC patients who cannot tolerate concurrent chemoradiation or are poor surgical candidates, pembrolizumab has shown to be superior to chemotherapy alone [59] and is now FDA-approved for this indication.

## BIOMARKERS

**PD-L1.** As PD-L1 is often expressed on tumor cells and binds to the PD-1 receptor, it was the first biomarker tested to predict the efficacy of the anti-PD1 therapies. Levels of tumor PD-L1 TPS by immunohistochemistry (IHC) expression correlated with levels of response to pembrolizumab [54]. The companion DAKO 22C3 PharmaDx IHC assay has been FDA-approved for use with pembrolizumab. In contrast, no correlation was identified between PD-L1 expression on tumors and response to nivolumab [53, 63] or atezolizumab [55]. The companion PD-L1 IHC assays differed for all antibodies. Merck utilized the DAKO 22C3 PharmaDx for pembrolizumab, BMS used DAKO 28-8 assay for nivolumab, Genentech employed Ventana SP142 assay for atezolizumab, and Medimmune/Astra Zeneca utilized the Ventana SP263 assay for durvalumab. The various antibodies used for IHC detection of PD-L1 can have different outcomes from each other (Fig. 6) [67, 68]. The Blueprint PD-L1 IHC Assay Comparison Project compared all four assays on 39 NSCLC tissues in the first phase [68]. 22C3 (pembrolizumab), 28-8 (nivolumab) and SP263 (durvalumab) correlated with each other for PD-L1 staining in tumor cells whereas the SP142 (atezolizumab) was clearly distinct. For immune cells, all four assays roughly correlated with each other but there was still large variability among the tests within an individual case. The phase 2 portion

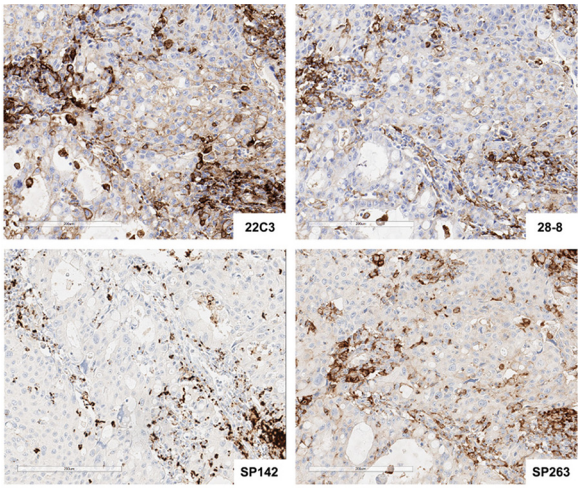



**Figure 6. PD-L1 staining by four different assays.** IHC stains for PD-L1 by assays used for clinical trials of immune checkpoint inhibitors on sections from same NSCLC sample. *From Hirsch et al., JTO, 2017.*

of the Blueprint project with analyses on larger number of cases and interpreting pathologists is underway.

**Total Mutation Burden.** Lung cancers contain some of the highest prevalence of mutations amongst all cancers due to cigarette smoke exposure [69]. The neoantigen theory posits that cancers with high mutation levels, such as melanoma and lung cancer, contain novel proteins ("neoantigens") as a consequence of these mutations that can act as T cell epitopes. A correlation between nonsynonymous mutation levels, as determined by whole exome sequencing, and response to pembrolizumab therapy was identified in NSCLC in a retrospective study [70]. Response rates were higher in those with high nonsynonymous tumor mutation burden (TMB, mutations > 200) compared to those with low TMB (<200) with significantly longer progression free survival. When nivolumab monotherapy was tested as first line for nonsquamous NSCLC [63], high TMB (>242 mutations) correlated with greater response and progression-free survival. No significant association between TMB and PD-L1 expression were identified.

DNA mismatch-repair deficiency (dMMR) leads to a large number of mutations including those in repetitive regions of DNA (microsatellite instability) [71]. A phase 2 trial assessed mismatch repair status with the efficacy of pembrolizumab as second line therapy for patients with metastatic carcinomas [72]. Colorectal cancer patients constituted two-thirds of the patients studied. dMMR colorectal and non-colorectal cancer patients showed substantially improved outcomes in response rate, overall and progression-free survival compared to mismatch repair-proficient (pMMR) patients. dMMR tumors averaged 1782 mutations per tumor compared to 73 mutations per tumor for pMMR tumors. FDA approved pembrolizumab for metastatic carcinomas with microsatellite instability-high or dMMR, irrespective of tumor type.

In contrast to the positive studies above, a recent phase III trial that compared nivolumab with ipilimumab, nivolumab alone and chemotherapy alone in metastatic NSCLC showed no correlation between high (>10 mutations per megabase) or low (<10 mutations per megabase) total mutation burden and overall survival [64].

## IMMUNE-RELATED ADVERSE EVENTS

Immune-related adverse events (IrAEs) can occur in virtually any organ (Fig. 7), often leading to a multidisciplinary management of the patient. CTLA-4 acts in the early stages of T-cell activation as the ligands for CTLA-4 are found almost exclusively on APCs whereas PD-1 activity occurs at later stages of T cell development. This may explain the distinct toxicity profiles of anti-CTLA-4 and anti-PD-1/PD-L1 therapies with greater incidence and severity of IrAEs with anti-CTLA-4 therapy [73]. After fatigue, skin rash and pruritis are the most common adverse events for both CTLA-4 and PD-1/L1 blockade. Colitis and hypophysitis occur more frequently with anti-CTLA-4 therapy whereas pneumonitis, hypothyroidism/thyroiditis, and arthralgias occur more frequently with anti-PD-1/L1 therapy [74]. Fatal tox-

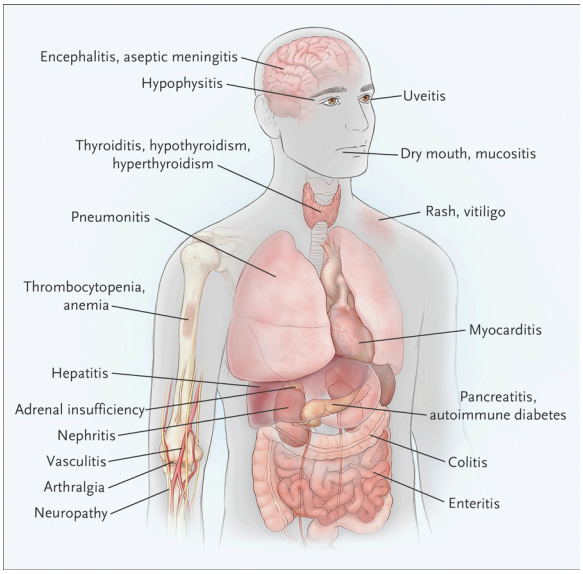



**Figure 7. Common adverse events from immune checkpoint blockade.** *From Postow et al., NEJM, 2018.*

ic events from pneumonitis, colitis, myocarditis, hepatitis and others have been reported [75].

IrAEs usually develop within the first several weeks to months after initiation of therapy. However, late incidences of IrAEs, even after cessation of therapy, have been reported [76]. Dermatologic events usually occur early in therapeutic course, followed by diarrhea/colitis several weeks later [76].

Specific treatments of IrAEs vary by the organ system involved. Although studies to support definitive therapy are lacking, guidelines for treatment of organ-specific IrAEs have been published [77, 78]. In general, for grade 1 toxicities, immune checkpoint therapy is continued with close monitoring. Immune checkpoint therapy is held for grade 2 toxicities until symptoms and/or lab values resolve. Prednisone 0.5 – 1.0 mg/kg/day may be initiated if the toxicity is not resolved within 1 week. For grade 3 immune checkpoint therapy is held and prednisone or methylprednisolone 1 – 2 mg/kg/day is initiated. Steroids can be slowly tapered over 4-6 weeks once toxicities subside to grade 1. If toxicities do not respond within 2 – 3 days, then infliximab or other alternative immune-suppressants are initiated. If symptoms do not improve in 4-6 weeks, checkpoint therapy is permanently discontinued. Management of grade 4 toxicities are similar except that immune checkpoint therapy is permanently discontinued. Life-threatening adverse event is an absolute contraindication for re-initiation of immune checkpoint therapy.

For NSCLC patients with IrAEs requiring discontinuation of anti-PD-1/L1 therapy, 26% of patients had a recurrent IrAE, 26% had a new IrAE and 48% had no IrAE with mostly mild second IrAEs [79]. Among patients who had no objective response prior to the first IrAE, re-initiation of anti-PD-1/L1 therapy led to longer overall and progression free survival [79]. For patients who had objective responses prior to the first IrAE, overall and progression-free survival were similar regardless of re-initiation or permanent discontinuation of anti-PD-1/L1 therapy [79].

**CONCLUSIONS**

Despite the gains made thus far, there are still many outstanding issues. The best response rates thus far are ~50% in patients treated with pembrolizumab and chemotherapy. Clearly, further research into basic biology of cancers and their interactions with the immune system is required. The difficulties with identification of a predictive biomarker for immune checkpoint therapy highlight the gaps in our knowledge and the need for standards in biomarker assays. The role of other components of the immune system, such as macrophages, neutrophils, NK cells, and STING pathway in tumor immunity is an area of intense preclinical study. Clinical trials with antagonists of other inhibitory immune checkpoints (such as TIM-3) and agonists of stimulatory immune checkpoints (such as OX-40) are underway. Also, clinical studies that combine radiation with immune checkpoint therapies have been initiated. Radiation-induced cell death can increase the repertoire of tumor antigens presented to T cells and thus, can potentially increase response rates and prolong survival of cancer patients on immune checkpoint therapies. The roles of immune checkpoint inhibitors in early stage lung cancers are also being explored. Finally, as immune checkpoint therapies become the new standard of care for multiple tumor types, studies are needed to optimize the treatment options for IrAEs.

Immune checkpoint therapy has transformed the landscape of lung cancer therapy and has become the new standard of therapy for all lung cancers that do not have targetable mutations in the locally advanced and advanced settings. The prolonged responses observed in a

subset of treated patients have been remarkable and unprecedented for stage IV disease, far surpassing the results with traditional chemotherapy and even targeted therapies. Current and future preclinical and clinical studies hold the promise of even further improvements in durable responses for lung cancer patients.

## REFERENCES


1. Siegel, R.L., K.D. Miller, and A. Jemal, *Cancer statistics, 2019.* CA Cancer J Clin, 2019. **69**(1): p. 7-34.

2. Siegel, R., D. Naishadham, and A. Jemal, *Cancer statistics, 2013.* CA: A Cancer Journal for Clinicians, 2013. **63**(1): p. 11-30.

3. National Cancer Institute Surveillance, E., and End Results Program. *Cancer Stat Facts: Lung and Bronchus Cancer*. Available from: https://seer.cancer.gov/statfacts/html/lungb.html.

4. Subramanian, J. and R. Govindan, *Lung cancer in never smokers: a review.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2007. **25**(5): p. 561-70.

5. *Chemotherapy in non-small cell lung cancer: a meta-analysis using updated data on individual patients from 52 randomised clinical trials. Non-small Cell Lung Cancer Collaborative Group.* BMJ, 1995. **311**(7010): p. 899-909.

6. Schiller, J.H., et al., *Comparison of four chemotherapy regimens for advanced non-small-cell lung cancer.* The New England Journal of Medicine, 2002. **346**(2): p. 92-8.

7. Scagliotti, G.V., et al., *Phase III study comparing cisplatin plus gemcitabine with cisplatin plus pemetrexed in chemotherapy-naive patients with advanced-stage non-small-cell lung cancer.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2008. **26**(21): p. 3543-51.

8. Johnson, D.H., et al., *Randomized phase II trial comparing bevacizumab plus carboplatin and paclitaxel with carboplatin and paclitaxel alone in previously untreated locally advanced or metastatic non-small-cell lung cancer.* J Clin Oncol, 2004. **22**(11): p. 2184-91.

9. Sandler, A., et al., *Paclitaxel-carboplatin alone or with bevacizumab for non-small-cell lung cancer.* N Engl J Med, 2006. **355**(24): p. 2542-50.

10. Lynch, T.J., et al., *Activating mutations in the epidermal growth factor receptor underlying responsiveness of non-small-cell lung cancer to gefitinib.* The New England Journal of Medicine, 2004. **350**(21): p. 2129-39.

11. Paez, J.G., et al., *EGFR mutations in lung cancer: correlation with clinical response to gefitinib therapy.* Science, 2004. **304**(5676): p. 1497-500.

12. Pao, W., et al., *EGF receptor gene mutations are common in lung cancers from "never smokers" and are associated with sensitivity of tumors to gefitinib and erlotinib.* Proceedings of the National Academy of Sciences of the United States of America, 2004. **101**(36): p. 13306-11.

13. Maemondo, M., et al., *Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR.* The New England Journal of Medicine, 2010. **362**(25): p. 2380-8.

14. Mitsudomi, T., et al., *Gefitinib versus cisplatin plus docetaxel in patients with non-small-cell lung cancer harbouring mutations of the epidermal growth factor receptor (WJTOG3405): an open label, randomised phase 3 trial.* The lancet oncology, 2010. **11**(2): p. 121-8.

15. Mok, T.S., et al., *Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma.* The New England Journal of Medicine, 2009. **361**(10): p. 947-57.

16. Rosell, R., et al., *Erlotinib versus standard chemotherapy as first-line treatment for European patients with advanced EGFR mutation-positive non-small-cell lung cancer (EURTAC): a multicentre, open-label, randomised phase 3 trial.* The lancet oncology, 2012. **13**(3): p. 239-46.

17. Zhou, C., et al., *Erlotinib versus chemotherapy as first-line treatment for patients with advanced EGFR mutation-positive non-small-cell lung cancer (OPTIMAL, CTONG-0802): a multicentre, open-label, randomised, phase 3 study.* The lancet oncology, 2011. **12**(8): p. 735-42.

18. Giaccone, G., et al., *Gefitinib in combination with gemcitabine and cisplatin in advanced non-small-cell lung cancer: a phase III trial--INTACT 1.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2004. **22**(5): p. 777-84.

19. Herbst, R.S., et al., *Gefitinib in combination with paclitaxel and carboplatin in advanced non-small-cell lung cancer: a phase III trial--INTACT 2.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2004. **22**(5): p. 785-94.

20. Gatzemeier, U., et al., *Phase III study of erlotinib in combination with cisplatin and gemcitabine in advanced non-small-cell lung cancer: the Tarceva Lung Cancer Investigation Trial.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2007. **25**(12): p. 1545-52.

21. Herbst, R.S., et al., *TRIBUTE: a phase III trial of erlotinib hydrochloride (OSI-774) combined with carboplatin and paclitaxel chemotherapy in advanced non-small-cell lung cancer.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2005. **23**(25): p. 5892-9.

22. Ohashi, K., et al., *Epidermal growth factor receptor tyrosine kinase inhibitor-resistant disease.* Journal of clinical oncology : official journal of the American Society of Clinical Oncology, 2013. **31**(8): p. 1070-80.

23. Finlay, M.R., et al., *Discovery of a potent and selective EGFR inhibitor (AZD9291) of both sensitizing and T790M resistance mutations that spares the wild type form of the receptor.* J Med Chem, 2014. **57**(20): p. 8249-67.

24. Cross, D.A., et al., *AZD9291, an irreversible EGFR TKI, overcomes T790M-mediated resistance to EGFR inhibitors in lung cancer.* Cancer Discov, 2014. **4**(9): p. 1046-61.

25. Soria, J.C., et al., *Osimertinib in Untreated EGFR-Mutated Advanced Non-Small-Cell Lung Cancer.* N Engl J Med, 2018. **378**(2): p. 113-125.

26. Mok, T.S., et al., *Osimertinib or Platinum-Pemetrexed in EGFR T790M-Positive Lung Cancer.* N Engl J Med, 2017. **376**(7): p. 629-640.

27. Soda, M., et al., *Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer.* Nature, 2007. **448**(7153): p. 561-6.

28. Hallberg, B. and R.H. Palmer, *Mechanistic insight into ALK receptor tyrosine kinase in human cancer biology.* Nature reviews. Cancer, 2013. **13**(10): p. 685-700.

29. Kwak, E.L., et al., *Anaplastic lymphoma kinase inhibition in non-small-cell lung cancer.* The New England Journal of Medicine, 2010. **363**(18): p. 1693-703.

30. Shaw, A.T., et al., *Crizotinib versus chemotherapy in advanced ALK-positive lung cancer.* The New England Journal of Medicine, 2013. **368**(25): p. 2385-94.

31. Hida, T., et al., *Alectinib versus crizotinib in patients with ALK-positive non-small-cell lung cancer (J-ALEX): an open-label, randomised phase 3 trial.* Lancet, 2017. **390**(10089): p. 29-39.

32. Peters, S., et al., *Alectinib versus Crizotinib in Untreated ALK-Positive Non-Small-Cell Lung Cancer.* N Engl J Med, 2017. **377**(9): p. 829-838.

33. Camidge, D.R., et al., *Brigatinib versus Crizotinib in ALK-Positive Non-Small-Cell Lung Cancer.* N Engl J Med, 2018. **379**(21): p. 2027-2039.

34. Novello, S., et al., *Alectinib versus chemotherapy in crizotinib-pretreated anaplastic lymphoma kinase (ALK)-positive non-small-cell lung cancer: results from the phase III ALUR study.* Ann Oncol, 2018. **29**(6): p. 1409-1416.

35. Kim, D.W., et al., *Brigatinib in Patients With Crizotinib-Refractory Anaplastic Lymphoma Kinase-Positive Non-Small-Cell Lung Cancer: A Randomized, Multicenter Phase II Trial.* J Clin Oncol, 2017. **35**(22): p. 2490-2498.

36. Shaw, A.T., et al., *Ceritinib versus chemotherapy in patients with ALK-rearranged non-small-cell lung cancer previously given chemotherapy and crizotinib (ASCEND-5): a randomised, controlled, open-label, phase 3 trial.* Lancet Oncol, 2017. **18**(7): p. 874-886.

37. Zou, H.Y., et al., *PF-06463922, an ALK/ROS1 Inhibitor, Overcomes Resistance to First and Second Generation ALK Inhibitors in Preclinical Models.* Cancer Cell, 2015. **28**(1): p. 70-81.

38. Gainor, J.F., et al., *Molecular Mechanisms of Resistance to First- and Second-Generation ALK Inhibitors in ALK-Rearranged Lung Cancer.* Cancer Discov, 2016. **6**(10): p. 1118-1133.

39. Shaw, A.T., et al., *Crizotinib in ROS1-rearranged non-small-cell lung cancer.* N Engl J Med, 2014. **371**(21): p. 1963-71.

40. Drilon, A., et al., *Safety and Antitumor Activity of the Multitargeted Pan-TRK, ROS1, and ALK Inhibitor Entrectinib: Combined Results from Two Phase I Trials (ALKA-372-001 and STARTRK-1).* Cancer Discov, 2017. **7**(4): p. 400-409.

41. Pardoll, D.M., *The blockade of immune checkpoints in cancer immunotherapy.* Nature reviews. Cancer, 2012. **12**(4): p. 252-64.

42. Brunet, J.F., et al., *A new member of the immunoglobulin superfamily--CTLA-4.* Nature, 1987. **328**(6127): p. 267-70.

43. Waterhouse, P., et al., *Lymphoproliferative disorders with early lethality in mice deficient in Ctla-4.* Science, 1995. **270**(5238): p. 985-8.

44. Kearney, E.R., et al., *Antigen-dependent clonal expansion of a trace population of antigen-specific CD4+ T cells in vivo is dependent on CD28 costimulation and inhibited by CTLA-4.* J Immunol, 1995. **155**(3): p. 1032-6.

45. Krummel, M.F., T.J. Sullivan, and J.P. Allison, *Superantigen responses and co-stimulation: CD28 and CTLA-4 have opposing effects on T cell expansion in vitro and in vivo.* Int Immunol, 1996. **8**(4): p. 519-23.

46. Leach, D.R., M.F. Krummel, and J.P. Allison, *Enhancement of antitumor immunity by CTLA-4 blockade.* Science, 1996. **271**(5256): p. 1734-6.

47. Ishida, Y., et al., *Induced expression of PD-1, a novel member of the immunoglobulin gene superfamily, upon programmed cell death.* EMBO J, 1992. **11**(11): p. 3887-95.

48. Nishimura, H., et al., *Development of lupus-like autoimmune diseases by disruption of the PD-1 gene encoding an ITIM motif-carrying immunoreceptor.* Immunity, 1999. **11**(2): p. 141-51.

49. Topalian, S.L., et al., *Safety, activity, and immune correlates of anti-PD-1 antibody in cancer.* The New England Journal of Medicine, 2012. **366**(26): p. 2443-54.

50. Brahmer, J.R., et al., *Safety and activity of anti-PD-L1 antibody in patients with advanced cancer.* The New England Journal of Medicine, 2012. **366**(26): p. 2455-65.

51. Topalian, S.L., et al., *Five-Year Survival and Correlates Among Patients With Advanced Melanoma, Renal Cell Carcinoma, or Non-Small Cell Lung Cancer Treated With Nivolumab.* JAMA Oncol, 2019.

52. Borghaei, H., et al., *Nivolumab versus Docetaxel in Advanced Nonsquamous Non-Small-Cell Lung Cancer.* N Engl J Med, 2015. **373**(17): p. 1627-39.

53. Brahmer, J., et al., *Nivolumab versus Docetaxel in Advanced Squamous-Cell Non-Small-Cell Lung Cancer.* N Engl J Med, 2015. **373**(2): p. 123-35.

54. Herbst, R.S., 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): p. 1540-50.

55. Rittmeyer, A., 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): p. 255-265.

56. Gettinger, S.N., 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): p. 2004-12.

57. Antonia, S.J., et al., *Nivolumab alone and nivolumab plus ipilimumab in recurrent small-cell lung cancer (CheckMate 032): a multicentre, open-label, phase 1/2 trial.* Lancet Oncol, 2016. **17**(7): p. 883-895.

58. Reck, M., et al., *Pembrolizumab versus Chemotherapy for PD-L1-Positive Non-Small-Cell Lung Cancer.* N Engl J Med, 2016. **375**(19): p. 1823-1833.

59. Mok, T.S.K., 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): p. 1819-1830.

60. Gandhi, L., et al., *Pembrolizumab plus Chemotherapy in Metastatic Non-Small-Cell Lung Cancer.* N Engl J Med, 2018.

61. Paz-Ares, L., et al., *Pembrolizumab plus Chemotherapy for Squamous Non-Small-Cell Lung Cancer.* N Engl J Med, 2018. **379**(21): p. 2040-2051.

62. Socinski, M.A., et al., *Atezolizumab for First-Line Treatment of Metastatic Nonsquamous NSCLC.* N Engl J Med, 2018. **378**(24): p. 2288-2301.

63. Carbone, D.P., et al., *First-Line Nivolumab in Stage IV or Recurrent Non-Small-Cell Lung Cancer.* N Engl J Med, 2017. **376**(25): p. 2415-2426.

64. Hellmann, M.D., et al., *Nivolumab plus Ipilimumab in Advanced Non-Small-Cell Lung Cancer.* N Engl J Med, 2019.

65. Horn, L., et al., *First-Line Atezolizumab plus Chemotherapy in Extensive-Stage Small-Cell Lung Cancer.* N Engl J Med, 2018. **379**(23): p. 2220-2229.

66. Antonia, S.J., et al., *Durvalumab after Chemoradiotherapy in Stage III Non-Small-Cell Lung Cancer.* N Engl J Med, 2017. **377**(20): p. 1919-1929.

67. Shukuya, T. and D.P. Carbone, *Predictive Markers for the Efficacy of Anti-PD-1/PD-L1 Antibodies in Lung Cancer.* J Thorac Oncol, 2016. **11**(7): p. 976-88.

68. Hirsch, F.R., 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): p. 208-222.

69. Alexandrov, L.B., et al., *Signatures of mutational processes in human cancer.* Nature, 2013. **500**(7463): p. 415-21.

70. Rizvi, N.A., et al., *Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer.* Science, 2015. **348**(6230): p. 124-8.

71. Poulogiannis, G., I.M. Frayling, and M.J. Arends, *DNA mismatch repair deficiency in sporadic colorectal cancer and Lynch syndrome.* Histopathology, 2010. **56**(2): p. 167-79.

72. Le, D.T., et al., *PD-1 Blockade in Tumors with Mismatch-Repair Deficiency.* N Engl J Med, 2015. **372**(26): p. 2509-20.

73. Michot, J.M., et al., *Immune-related adverse events with immune checkpoint blockade: a com-*

*prehensive review.* Eur J Cancer, 2016. **54**: p. 139-148.

74. Postow, M.A., R. Sidlow, and M.D. Hellmann, *Immune-Related Adverse Events Associated with Immune Checkpoint Blockade.* N Engl J Med, 2018. **378**(2): p. 158-168.

75. Wang, D.Y., et al., *Fatal Toxic Effects Associated With Immune Checkpoint Inhibitors: A Systematic Review and Meta-analysis.* JAMA Oncol, 2018. **4**(12): p. 1721-1728.

76. Weber, J.S., et al., *Safety Profile of Nivolumab Monotherapy: A Pooled Analysis of Patients With Advanced Melanoma.* J Clin Oncol, 2017. **35**(7): p. 785-792.

77. Puzanov, I., et al., *Managing toxicities associated with immune checkpoint inhibitors: consensus recommendations from the Society for Immunotherapy of Cancer (SITC) Toxicity Management Working Group.* J Immunother Cancer, 2017. **5**(1): p. 95.

78. Brahmer, J.R., et al., *Management of Immune-Related Adverse Events in Patients Treated With Immune Checkpoint Inhibitor Therapy: American Society of Clinical Oncology Clinical Practice Guideline.* J Clin Oncol, 2018. **36**(17): p. 1714-1768.

79. Santini, F.C., et al., *Safety and Efficacy of Re-treating with Immunotherapy after Immune-Related Adverse Events in Patients with NSCLC.* Cancer Immunol Res, 2018. **6**(9): p. 1093-1099.