Available online at www.sciencedirect.com

ScienceDirect

Journal of the Chinese Medical Association 80 (2017) 7–14

www.jcma-online.com

Review Article

Immune checkpoint inhibitors for nonsmall cell lung cancer treatment

Yuh-Min Chen [a,b,c,*]

[a] *Division of General Chest Medicine, Department of Chest Medicine, Taipei Veterans General Hospital, Taipei, Taiwan, ROC*
[b] *School of Medicine, National Yang-Ming Medical University, Taipei, Taiwan, ROC*
[c] *School of Medicine, Taipei Medical University, Taipei, Taiwan, ROC*

Received February 21, 2016; accepted July 18, 2016

Abstract

Immune checkpoint inhibition with blocking antibodies that target cytotoxic T-lymphocyte antigen-4 (CTLA-4) and the programmed cell death protein 1 (PD-1) pathway [PD-1/programmed death-ligand 1 (PD-L1)] have demonstrated promise in a variety of malignancies. While ipilimumab has been approved as a CTLA-4 blocking antibody by the US Food and Drug Administration for the treatment of advanced melanoma, it is still not approved for lung cancer treatment. In contrast, nivolumab and pembrolizumab, both PD-1 blocking antibodies, have been approved for second-line treatment of nonsmall cell lung cancer in 2015 because of their high potency and long-lasting effects in some patient subgroups. Other PD-1 and PD-L1 monoclonal antibodies are also in active development phase. Treatment with such immune checkpoint inhibitors is associated with a unique pattern of immune-related adverse events or side effects. Combination approaches involving CTLA-4 and PD-1/PD-L1 blockade or checkpoint inhibitors with chemotherapy or radiotherapy are being investigated to determine whether they may enhance the efficacy of treatment. Despite many challenges ahead, immunotherapy with checkpoint inhibitors has already become a new and important treatment modality for lung cancer in the last decade following the discovery of targeted therapy.

Keywords: adenocarcinoma; checkpoint inhibitor; immunotherapy; lung cancer; lymphocytes

1. Introduction

Lung cancer is the leading cause of cancer-related deaths in Taiwan and other developed countries in the world. The 5-year survival rate was only 15.9%, with a median survival of 13.2 months, in Taiwan between 2002 and 2008.[1]

There are two arms of the immune system, the innate and the adaptive, which protect the body from foreign agents. The innate immune system includes physical epithelial barriers, phagocytes, natural killer cells, and circulating complement proteins. The innate immune system is the first line of defense against pathogens. In contrast, the adaptive arm of the immune system is dormant until it is primed by the presence of a pathogen that has evaded or overwhelmed the innate immunity. Components of the adaptive immune system include both B cells and T cells. Naïve B cells are activated to produce antigen-recognizing antibodies when they are presented with antigens from a pathogen. When foreign antigens are presented to naïve T cells, they mature into one of two types of effector T cells: $CD4^+$ helper T cells that facilitate antibody production, and $CD8^+$ cytotoxic T cells that directly kill cells recognized as foreign (such as viral infected cells or tumor cells); this process is called cell-mediated immunity. The adaptive immune response is initiated when tumor cell antigens released by innate immunity are taken up by dendritic cells. These dendritic cells then migrate to the draining lymph nodes, where they present these tumor antigens to T cells, causing them to mature into cytotoxic T cells to destroy tumor cells (Fig. 1).

Conflicts of interest: The author declares that he has no conflicts of interest related to the subject matter or materials discussed in this article.

* Corresponding author. Dr. Yuh-Min Chen, Department of Chest Medicine, Taipei Veterans General Hospital, 201, Section 2, Shih-Pai Road, Taipei 112, Taiwan, ROC.

E-mail address: ymchen@vghtpe.gov.tw.

http://dx.doi.org/10.1016/j.jcma.2016.08.005

Although the immune system plays an important role in recognizing, controlling, and eradicating cancer, cancer induces immunosuppression through several mechanisms that may suppress effective antitumor immunity, including but not limited to: (1) secretion of immunosuppressive cytokines; (2) loss of major histocompatibility complex antigen expression; (3) and programmed cell death protein 1/programmed cell death protein 1 ligand (PD-1/PD-L1) interaction of tumor cells with immune cells.[2−6] In the past, immunotherapy has had minimal success in lung cancer treatment, which was attributed in part to the belief that lung cancer is non-immunogenic.[7−11] Most patients present with advanced disease and are immunosuppressed, as documented by decreased lymphocyte counts and cytotoxic function seen in this patient population.[8,11−13] Regulatory T-cells ($CD4^+$ Treg) are a subpopulation of lymphocytes that play an important role in suppressing tumor immune surveillance, and have been found to have higher levels in peripheral blood and tumor microenvironment of lung cancer patients compared with other T-cell subpopulations.[14] $CD4^+$ Treg suppress cytotoxic T-cell functions that are responsible for killing tumor cells. We previously also showed that double signal stimulation is needed for these immunosuppressed lymphocytes to recover their cytotoxic function against tumor cells.[9,15−18]

It was recently found that cancer cells can prevent themselves from immune surveillance and killing through adaptive immune resistance, causing the disabling of tumor-specific T cells (Fig. 2).[19,20] Many types of cancers have been found to express PD-L1 on their tumor cell surfaces, which is a known ligand of the PD-1 receptor on T cells. This pathway of interaction between PD-1 and PD-L1 causes T-cell downregulation and functional inhibition.[5,6] There are two immune checkpoint inhibitory pathways that involve signaling through CTLA-4 or PD-1 with their ligands (Figs. 1 and 2). Antibody therapies against these negative immunologic regulators have demonstrated significant success in lung cancer treatment in recent years. This review focuses on antibodies that block the CTLA-4 and PD-1/PD-L1 pathways. We discuss the preclinical rationale and clinical experience with these antibodies in nonsmall cell lung cancer (NSCLC) treatment.

2. Cytotoxic T-lymphocyte antigen-4 pathway

Cytotoxic T-lymphocyte antigen-4 (CTLA-4) is a receptor that is expressed on the T-cell surface during the T-cell activation phase. Activation of the T-cell requires both antigen presentation in the context of a major histocompatibility complex molecule and a costimulatory signal stimulation by B7 from an antigen-presenting cell to interact with CD28 on the T-cell. Early after T-cell activation, CTLA-4 is translocated to the plasma membrane of the T-cell. CTLA-4 binds members of the B7 family with a much higher affinity than CD28, where it downregulates the function of activated T-cells (Fig. 1). CTLA-4 downregulates activated T-cell function not only through preventing costimulation by outcompeting CD28 for its ligand, B7, but also by inducing T-cell cycle arrest.[21−24] Through these mechanisms, CTLA-4 has an essential role in maintaining normal immunologic homeostasis, as evidenced by the fact that mice deficient in CTLA-4 died from fatal lymphoproliferative disease.[25]

CTLA-4 also regulates tumor immunity via Treg that expresses high levels of surface CTLA-4. CTLA-4-expressing

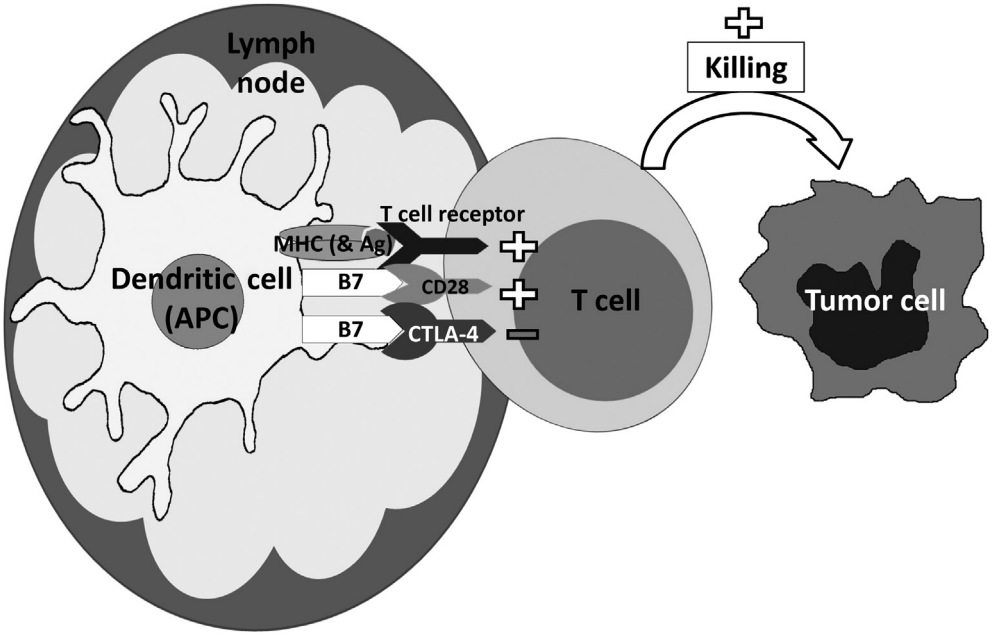

Fig. 1. T-cell activation phase and the cytotoxic T-lymphocyte antigen-4 (CTLA-4) immunologic checkpoint. T-cell activation requires antigen presentation in the context of a major histocompatibility complex (MHC) molecule in addition to the costimulatory signal stimulation when B7 on an antigen-presenting cell interacts with CD28 on a T cell. Soon after activation, CTLA-4 is translocated to the plasma membrane where it downregulates the function of T cells to maintain immunologic homeostasis.

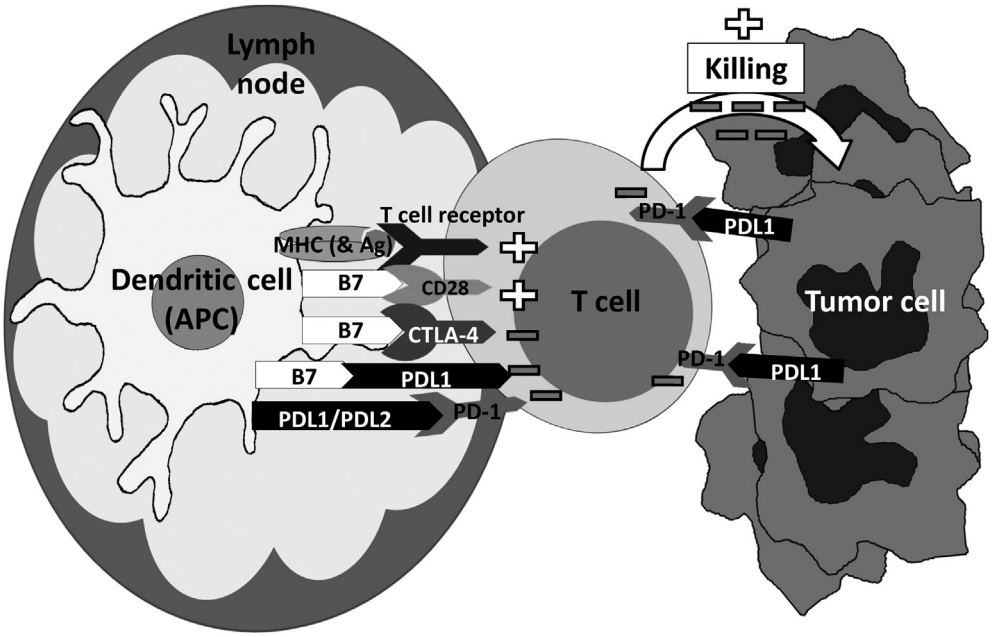

Fig. 2. T-cell effector phase and the programmed cell death protein 1 (PD-1) immunologic checkpoint. PD-1 is expressed on activated T cells. Interactions between PD-1 and its ligands, programmed death-ligand (PD-L)1 and PD-L2, are complex and occur at many steps of an immune response. An interaction soon after activation in the lymph node where PD-L1 or PD-L2 on an antigen-presenting cell negatively regulates T-cell activity through PD-1 and through an interaction between B7 and PD-L1. The PD-1 pathway is important in the tumor microenvironment, where PD-L1 expressed by tumors interacts with PD-1 on T cells to suppress T-cell effector function.

Tregs may facilitate nonresponsiveness of the immune system to tumor antigens.[26] Tregs have been shown to be present in tumors and coexist with primed effector T cells. Thus, blocking Treg function through anti-CTLA-4 antibodies may have the potential to remove Treg suppression and enhance antitumor immunity.

Based on preclinical studies that demonstrated antibody blockades of CTLA-4 could result in antitumor immunity,[27,28] two antibodies targeting CTLA-4, ipilimumab (Bristol-Myers Squibb, Princeton, NJ, USA) and tremelimumab (MedImmune/AstraZeneca, Wilmington, DE, USA) entered clinical development. Early reports on both agents showed durable clinical responses in some patients, particularly melanoma patients.[29–31] Ipilimumab is a fully human anti-CTLA-4 monoclonal antibody that is now approved for the treatment of melanoma.[32]

The rationale for the combined use of anti-CTLA-4 antibody with chemotherapy is based on the assumption that tumor-specific antigens will be released during chemotherapy-induced tumor necrosis and that will augment tumor-specific immune reactions. Ipilimumab plus chemotherapy showed promising results in a phase II CA184-041 study that randomized previously untreated advanced NSCLC patients to receive paclitaxel plus carboplatin, alone or in association with concurrent ipilimumab (10 mg/kg from Cycle 1 to Cycle 4) or with phased ipilimumab (10 mg/kg from Cycle 3 to Cycle 6).[33] There were 204 patients included in this study. Response was assessed by using immune-related response criteria and modified World Health Organization criteria.[34,35] This study met its primary end point of improved immune-related progression-free survival (PFS) for phased ipilimumab versus the control [hazard ratio (HR) = 0.72, $p = 0.05$], but not for concurrent ipilimumab (HR = 0.81, $p = 0.13$). For the phased arm, concurrent arm, and control arm, the median immune-related PFSs were 5.7 months, 5.5 months, and 4.6 months, respectively; median PFSs of 5.1 months, 4.1 months, and 4.2 months, respectively; immune-related best overall response rates of 32%, 21%, and 18%, respectively; and best overall response rates of 32%, 21%, and 14%, respectively.

A phase III study of paclitaxel/carboplatin with or without ipilimumab in treatment-naive squamous NSCLC is ongoing.

Another CTLA-4-blocking antibody, tremelimumab (CP-675,206) is a fully human immunoglobulin G (IgG)-2 monoclonal antibody. Tremelimumab has induced durable tumor responses in patients with melanoma in a phase I/II clinical trial.[36] However, a phase III trial was discontinued after review of interim data showed that the trial would not demonstrate superiority to conventional standard chemotherapy.[37] Tremelimumab has shown promising responses in patients with malignant mesothelioma.[38]

3. PD-1/PD-L1 pathway

Successful treatment targeting CTLA-4 has created enthusiasm for approaches targeting other immunologic checkpoints. Among them, the PD-1/PD-L1 axis has been most actively studied (Figure 2). PD-1 is a negative regulator of T-cell activity that limits the activity of T cells, especially in the effector phase, when it interacts with its two ligands PD-L1 and PD-L2.[5,39,40] When engaged by the ligand, PD-1

inhibits kinase signaling pathways that normally lead to T-cell activation.[5] Mice that are deficient in PD-1 have a different and distinct autoimmune phenotype from mice deficient in CTLA-4.[41,42] PD-1 is expressed on many types of lymphocytes, including B cells and natural killer cells.[40,43]

There are several antibodies that disrupt the PD-1 axis that have entered clinical development; two of them (nivolumab, Bristol-Myers Squibb, New York, NY, USA; pembrolizumab, Merck, Whitehouse Station, NJ, USA) have Food and Drug Administration (FDA) approval for second-line treatment of NSCLC (Table 1). These antibodies can be classified into two main categories: those that target PD-1 and those that target PD-L1.

3.1. Nivolumab

Nivolumab (ONO-4538, BMS-936558) is a human IgG4 monoclonal antibody that targets the PD-1 receptor.[44−47] A report of the long-term follow-up of 129 patients with heavily pretreated NSCLC who entered a phase I dose-escalation cohort expansion trial of nivolumab 1 mg/kg, 3 mg/kg, or 10 mg/kg intravenously (IV) once every 2 weeks revealed that 1-, 2-, and 3-year overall survival (OS) rates were 42%, 24%, and 18%, respectively, across doses, and were 56%, 42%, and 27%, respectively, at the 3 mg/kg dose ($n = 37$) chosen for further clinical development.[45] Response rates were similar in squamous (16.7%) and nonsquamous (17.6%) NSCLC. A phase II trial of squamous type NSCLC was performed in France, Germany, Italy, and the USA. Patients with squamous NSCLC who had received two or more previous treatments received IV nivolumab (3 mg/kg) every 2 weeks until progression or unacceptable toxic effects. Between 2012 and 2013, 117 patients were enrolled.[44] Seventeen (14.5%) of 117 patients had an objective response. The response rate was 14% in patients with tumor PD-L1 expression $< 5\%$ and 24% in those with expression $\geq 5\%$. A phase III trial was done on 272 squamous NSCLC patients who had disease progression during or after first line chemotherapy (CheckMate 017).[46] Patients were randomized to receive nivolumab 3 mg/kg every 2 weeks, or docetaxel 75 mg/m^2 every 3 weeks. The median OS was 9.2 months with nivolumab versus. 6.0 months with docetaxel (HR = 0.59, $p < 0.001$). One-year survival rate was 42% with nivolumab versus 24% with docetaxel. The response rate was 20% with nivolumab versus 9% with docetaxel ($p = 0.008$). The median PFS was 3.5 months with nivolumab versus 2.8 months with docetaxel (HR = 0.62, $p < 0.001$). The expression of the PD-L1 was neither prognostic nor predictive of treatment benefit. The US FDA granted approval to nivolumab for the treatment of metastatic squamous NSCLC patients with progression on or after platinum-based chemotherapy in 2015.

In CheckMate 057, patients with nonsquamous NSCLC that had progressed during or after platinum-based chemotherapy were randomized to receive nivolumab or docetaxel.[47] The median OS was 12.2 months among 292 patients in the nivolumab arm and 9.4 months among 290 patients in the docetaxel arm (HR = 0.73, $p = 0.002$). One-year survival rate was 51% with nivolumab versus 39% with docetaxel. The response rate was 19% with nivolumab versus 12% with docetaxel ($p = 0.02$). Based on these data, the FDA approved nivolumab for the treatment of metastatic NSCLC patients with progression on or after platinum-based chemotherapy recently. This approval expands the indication for nivolumab in NSCLC to include nonsquamous histologies.

A phase I study evaluating the efficacy and safety of nivolumab monotherapy in patients with chemotherapy naïve advanced NSCLC was reported recently.[48] There were 52 advanced NSCLC patients who received nivolumab 3 mg/kg IV every 2 weeks until disease progression or unacceptable toxicity. Preliminary results showed that the response rate (RR) was 21% (11/52). Objective RRs for subgroups were 23% (9/39) in nonsquamous and 15% (2/13) in squamous NSCLC. Objective responses were 31% (8/26) in PD-L1 positive patients and 10% (2/21) in PD-L1 negative patients.

Nivolumab has been combined with platinum-based chemotherapy or anti-CTLA4 immunotherapy as first-line treatment for advanced NSCLC, or with epidermal growth factor receptor (EGFR)-tyrosine kinase inhibitor targeted therapy for EGFR-tyrosine kinase inhibitor acquired resistance.[49−51] The data are too premature to be discussed.

3.2. Pembrolizumab

Pembrolizumab (MK-3475, lambrolizumab, Keytruda, Kenilworth, NJ, USA) is an IgG4-engineered humanized antibody that targets the PD-1 receptor. A recently published paper

Table 1
Immune checkpoint inhibitors in development for nonsmall cell lung cancer.

Inhibitor	Target	Monoclonal antibody type	Company	Development phase
Nivolumab (ONO-4538, BMS-936558)	PD-1	Fully human IgG4	Ono Pharmaceutical/Bristol-Myers Squibb	FDA-approved
Pembrolizumab (MK-3475)	PD-1	Humanized IgG4	Merck Sharp & Dohme	FDA-approved
Atezolizumab (MPDL3280A)	PD-L1	Human IgG1	Genentech/Roche	III
Durvalumab (MEDI-4736)	PD-L1	Fully human IgG1	MedImmune/Astra-Zeneca	III
Avelumab (MSB0010718C)	PD-L1	Fully human IgG1	Merck/Pfizer	III
Ipilimumab	CTLA-4	Fully human IgG1	Bristol-Myers Squibb	III
Tremelimumab (CP-675,206)	CTLA-4	Fully human IgG2	MedImmune/Pfizer	III

CTLA-4 = cytotoxic T-lymphocyte antigen-4; FDA = Food and Drug Administration; IgG = immunoglobulin G; PD-1 = programmed cell death protein 1; PD-L1 = programmed cell death-ligand 1

included 495 NSCLC patients who received pembrolizumab (at a dose of either 2 mg/kg or 10 mg/kg every 3 weeks or 10 mg/kg every 2 weeks) as part of the international phase 1 KEYNOTE-001 trial.[52] Among all the NSCLC patients, the objective RR was 19.4%, and the median duration of response was 12.5 months. The median duration of OS was 12.0 months. Among patients with a PD-L1 proportion score of at least 50%, the RR was 45.2% in the validation group. Among all the patients with a proportion score of at least 50%, median PFS was 6.3 months. The US FDA granted accelerated approval to pembrolizumab for metastatic NSCLC treatment in patients whose tumors expressed PD-L1, as determined by an FDA-approved test, with disease progression during or after platinum-containing chemotherapy in 2015.[53]

The KEYNOTE-021 trial evaluated the safety, tolerability, and clinical activity of pembrolizumab plus platinum-based doublet chemotherapy for treatment-naïve patients with advanced NSCLC.[54] Patients were randomized 1:1 to pembrolizumab 2 or 10 mg/kg every 3 weeks plus carboplatin and paclitaxel (Cohort A; any histology) or carboplatin plus pemetrexed (Cohort C; nonsquamous without *EGFR* sensitizing mutation nor *anaplastic lymphoma kinase* translocation). Patients received pembrolizumab plus chemotherapy for four cycles followed by pembrolizumab maintenance therapy in Cohort A and pembrolizumab plus pemetrexed maintenance therapy in Cohort C. As of December 2014, 44 patients were treated. Preliminary RR was 30% in Cohort A and 58% in Cohort C.

Since combined anti-PD-1 and anti-CTLA-4 treatment has shown robust efficacy and manageable toxicity in patients with melanoma, a phase 1 study evaluating pembrolizumab plus ipilimumab was performed in NSCLC patients.[55] Patients with NSCLC that recurred after no more than two prior regimens received pembrolizumab plus ipilimumab every 3 weeks for four cycles followed by maintenance pembrolizumab therapy. The preliminary data demonstrated an acceptable toxicity profile and robust antitumor activity for pembrolizumab plus ipilimumab in patients with recurrent NSCLC.

Regarding immunotherapy in NSCLC patients with brain metastases, a preliminary report of phase II pembrolizumab on patients with at least one brain metastasis that was previously untreated or progressing after prior local therapy showed promising results, with four of an initial nine patients who had brain metastatic lesion showing partial response to the treatment.[56] The study is still ongoing.

3.3. Atezolizumab

Atezolizumab (MPDL3280A) is a humanized, engineered monoclonal antibody of IgG1 against PD-L1. Preliminary results of two phase II trials in NSCLC were reported recently. One of the studies (POPLAR, $n = 287$) was a randomized trial with docetaxel as the control arm; the data showed that atezolizumab significantly improved OS.[57] In this study, previously treated NSCLC patients were randomized to receive atezolizumab 1200 mg IV every 3 weeks or docetaxel 75 mg/m^2 IV every 3 weeks. PD-L1 expression was evaluated using the SP142 antibody assay. Patients were grouped as tumor cell PD-L1 staining (TC) 0, 1, 2, or 3 and immune cell PD-L1 staining (IC) 0, 1, 2, or 3. Improved efficacy was found with increasing PD-L1 expression (OS: HR = 0.47; PFS: HR = 0.56; RR = 38% vs. 13% in TC3 or IC3 patients comparing atezolizumab with docetaxel), while patients with the lowest PD-L1 levels (TC0 and IC0) did not appear to benefit from atezolizumab (OS: HR = 1.22).[58] Median OS was 12.6 months with atezolizumab versus 9.7 months with docetaxel (HR = 0.73, $p = 0.04$). However, there was no difference between atezolizumab and docetaxel (median OS of 9.7 months in both arms) in patients with little or no expression of PD-L1. The other study (known as BIRCH) was a single-arm study in which atezolizumab was used in patients with PDL1-positive advanced NSCLC.[59] PD-L1 expression was assessed in the same way as in the POPLAR study. Patients were divided into three cohorts: Cohort 1 had no prior therapy, Cohort 2 had received one prior chemotherapy, and Cohort 3 had received at least two prior systemic therapies. The primary endpoint was RR, which was 26%, 24%, and 27%, respectively, for the three cohorts of patients who had a high expression of PD-L1. The response rates were 19%, 17%, and 17%, respectively, for patients who had a medium to high expression of PDL1. Another phase II study of atezolizumab (FIR) in stage IIIB/IV NSCLC patients based on PD-L1 expression was also reported recently.[60] Cohort 1 included chemonaive patients, Cohort 2 included patients who had received no less than two lines of systemic treatment without brain metastasis, and Cohort 3 included patients who had received no less than two lines of systemic treatment and were with treated asymptomatic brain metastasis. Atezolizumab dose and PD-L1 expression and scoring system were the same as in POPLAR and BIRCH. Patients with PD-L1 TC 2/3 and/or IC 2/3 tumors were enrolled. Of the 138 patients enrolled, the response rates were 29%, 17%, and 17% in Cohort 1, Cohort 2, and Cohort 3, respectively, while the highest response rates were seen in patients with PD-L1 TC3 or IC3 tumors (29%, 26%, and 25%, respectively). It seems that atezolizumab had a remarkable activity in NSCLC patients with high PD-L1 expression regardless of the line of treatment.

A phase Ib study that evaluated atezolizumab in combination with carboplatin plus either paclitaxel (Arm C), pemetrexed (Arm D) or weekly nab-paclitaxel (Arm E) was done in chemonaive locally advanced or metastatic NSCLC, and preliminary results are available.[61] Patients received atezolizumab every 3 weeks with a standard chemotherapy dosing for four to six cycles followed by atezolizumab maintenance therapy until disease progression. Across all arms, the RR was 67% (20 of 30), including 60% in Arm C (three of five), 75% in Arm D (eight of 12), and 62% in Arm E (eight of 13). Phase III studies are ongoing.

3.4. Durvalumab

Durvalumab (MEDI-4736) is a human IgG1 monoclonal antibody targeting PD-L1. Preliminary results of an ongoing

Table 2
Summary of programmed cell death-ligand 1 immunohistochemistry in nonsmall cell lung cancer (NSCLC) clinical trials.

Drug	Marker antibody	Treatment line	Definition of "positive", %	No. positive, %	Positive predictive outcome	RR%, overall	RR% IHC positive cases	RR% IHC negative cases	Ref.
Nivolumab	Dako 28-8	≥ 2nd, squamous	≥ 10, ≥ 5, ≥ 1 in > 100 cells	83 for ≥ 1	No	20	19, 21, 18	16, 15, 17	46
Nivolumab	Dako 28-8	≥ 2nd, nonsquamous	≥ 10, ≥ 5, ≥ 1 in > 100 cells	78 for ≥ 1	Yes	19	37, 36, 31	11, 10, 9	47
Pembrolizumab	Dako 22C3	Any line	≥ 50	23.2	Yes	19.4	45.2	NR	52
Atezolizumab (MPDL3280A)	Roche Ventana, SP142	2nd or 3rd, NSCLC	Tumor cell: ≥ 50, ≥ 5, ≥ 1 or immune cell: ≥ 10, ≥ 5, ≥ 1	68 for ≥ 1	Yes	15	38, 22, 18	8	57, 58
Durvalumab (MedI-4736)	Roche Ventana, SP142 Roche Ventana, SP263	1st (1L), 2nd (2L), or ≥ 3rd (3L+), NSCLC Any line, NSCLC	Tumor cell: ≥ 50, ≥ 5 or Immune cell: ≥ 10, ≥ 5 ≥ 25% of tumor cell at any intensity	Nil 48	Yes Yes	1L:19, 2L:17, 3L+:17 16	1L:19, 2L:17, 3L+:17 27	Nil 5	59 62
Avelumab (MSB0010718C)	Dako, clone: not reported	PD after 1 line of platinum -containing doublet c/t, NSCLC	≥ 1% of tumor cells at any intensity	66.3	Yes	13.6	15.6	10	63

IHC = immunohistochemistry; PD = programmed death; RR = response rate.

phase I/II, multicenter study of durvalumab evaluating the safety and clinical activity of durvalumab in patients with multiple solid tumor types including NSCLC were reported recently.[62] Durvalumab was administered at 10 mg/kg IV every 2 weeks until unacceptable toxicity, disease progression, or 12 months was reached. Tumor PD-L1 expression was assessed using Ventana PD-L1 IHC (SP263). As of October 31, 2014, 198 patients had been treated. There were 149 patients evaluable for response, and RR was 14% (23% in tumor PD-L1 positive patients). The response rate was higher in squamous (21%) than in nonsquamous patients (10%). Durvalumab has been accelerated into phase III clinical development in NSCLC at present.

3.5. Avelumab

Avelumab (MSB0010718C) is a fully human anti-PD-L1 IgG1 monoclonal antibody. By retaining a native Fc-region, avelumab is also able to induce antibody-dependent cell-mediated cytotoxicity.

There was a preliminary report of a phase Ib expansion trial evaluating safety and clinical activity in patients with advanced NSCLC progressing after platinum-based chemotherapy.[63] Patients were treated with avelumab at 10 mg/kg every 2 weeks until disease progression, confirmed complete response, or intolerable toxicity. A follow-up analysis of 184 patients was performed. Objective responses were observed in 22 (12%) patients. Median PFS was 2.7 months. The RR in PD-L1 positive patients ($n = 118$) was 14.4% and 10.0% in PD-L1 negative patients ($n = 20$).

4. Surrogate marker

Among predictors for checkpoint inhibitor therapy, tumor PD-L1 immunohistochemical staining is the most frequently used predictor for anti-PD-1 and anti-PD-L1 immunotherapy.[64,65] There are at least four kinds of kits or platforms for detection of tumor PD-L1 expression. Some studies also count PD-L1 expression in immune cells and correlate both tumor and immune cell PD-L1 expression with treatment response. Most studies assess PD-L1 expression in tumor cells and regard membrane staining as most significant.[46,47,52,64] In general, there is a trend of higher response rates in the PD-L1 expression positive patients compared with the PD-L1 expression negative patients, although in some studies this difference was not significant (Table 2). Up to now, PD-L1 expression is not a perfect biomarker, since most studies also report significant response rates (3−20%) in PD-L1 expression negative patients.[65] Other proposed methodologies include examination of mutational burden by genomics.[66]

In conclusion, with the approval of both nivolumab and pembrolizumab in the treatment of second-line NSCLC treatment, the use of immune checkpoint inhibitors for the treatment of NSCLC is firmly established. The ongoing plethora of phase III studies of PD-1 and PD-L1 inhibitors, either alone or in combination with chemotherapy, targeted

therapy, radiotherapy, or immunotherapy in different stages of NSCLC, will serve to clarify and likely expand their use in NSCLC treatment. However, there are still many challenges ahead for oncologists, including determining the optimal time for integration of immunotherapy into the lifetime course of NSCLC patient treatment. Despite these challenges, immunotherapy with checkpoint inhibitor has already become the newest, most important, and most novel treatment modality during the decade following the discovery of targeted therapy.

References

1. Wang BY, Huang JY, Cheng CY, Lin CH, Ko J, Liaw YP. Lung cancer and prognosis in Taiwan: a population-based cancer registry. *J Thorac Oncol* 2013;**8**:1128−35.
2. Dasanu CA, Sethi N, Ahmed N. Immune alterations and emerging immunotherapeutic approaches in lung cancer. *Expert Opin Biol Ther* 2012;**12**:923−37.
3. Marincola FM, Jaffee EM, Hicklin DJ, Ferrone S. Escape of human solid tumors from T-cell recognition: molecular mechanisms and functional significance. *Adv Immunol* 2000;**74**:181−273.
4. Brahmer JR. Harnessing the immune system for the treatment of non-small-cell lung cancer. *J Clin Oncol* 2013;**31**:1021−8.
5. Freeman GJ, Long AJ, Iwai Y, Bourque K, Chernova T, Nishimura H, et al. Engagement of the PD-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. *J Exp Med* 2000;**192**:1027−34.
6. Chen L. Co-inhibitory molecules of the B7-CD28 family in the control of T-cell immunity. *Nat Rev Immunol* 2004;**4**:336−47.
7. Holt GE, Podack ER, Raez LE. Immunotherapy as a strategy for the treatment of non-small-cell lung cancer. *Therapy* 2011;**8**:43−54.
8. Chen YM, Yang WK, Ting CC, Tsai WY, Yang DM, Whang-Peng J, et al. Cross regulation by IL-10 and IL-2/IL-12 of the cytolytic activity of lymphocyte from malignant effusion of lung cancer patients. *Chest* 1997;**112**:960−6.
9. Chen YM, Yang WK, Whang-Peng J, Tsai WY, Hung YM, Yang DM, et al. Restoration of the immunocompetence by IL-2 activation and TCR-CD3 engagement of the in vivo anergized tumor specific CTL from lung cancer patients. *J Immunotherapy* 1997;**20**:354−64.
10. Chen YM, Yang WK, Whang-Peng J, Kuo BIT, Perng RP. Elevation of interleukin-10 levels in malignant pleural effusion. *Chest* 1996;**110**:433−6.
11. Chen YM, Whang-Peng J, Yang WK, Hung YM, Lin WC, Kuo BIT, et al. Low levels of NK cells and related cytokines in pleural effusion. *J Chin Med Assoc* 1996;**58**:156−62.
12. Chen YM, Yang WK, Yang KY, Whang-Peng J, Tsai CM, Perng RP. An analysis of cytokine status in the serum and effusions of patients with tuberculous and lung cancer. *Lung Cancer* 2001;**31**:25−30.
13. Wesselius LJ, David L, Wheaton BS, Wahl LJM, Sherad S, Taylor SA. Lymphocyte subsets in lung cancer. *Chest* 1987;**91**:725−9.
14. Woo EY, Yeh H, Chu CS, Schlienger K, Carroll RG, Riley JL, et al. Cutting edge: Regulatory T cells from lung cancer patients directly inhibit autologous T cell proliferation. *J Immunol* 2002;**168**:4272−6.
15. Chen YM, Tsai CM, Whang-Peng J, Perng RP. Double signal stimulation was required for full recovery of the autologous tumor-killing effect of effusion-associated lymphocytes. *Chest* 2002;**122**:1421−7.
16. Chen YM, Tsai CM, Perng RP. Differential effects of different cytokines on the tumorigenicity and immunogenicity of murine tumors. *J Chin Med Assoc* 1999;**62**:807−16.
17. Chen YM, Ting CC, Whang-Peng J, Yang KY, Yang WK, Tsai CM, et al. Restoration of cytotoxic T lymphocytes function in malignant pleural effusion: interleukin-15 versus interleukin-2. *J Interferon Cytokine Res* 2000;**20**:31−9.
18. Chen YM, Tsai CM, Whang-Peng J, Perng RP. IL-7 and IL-12 have different effects in recovery of depressed cellular immunity of malignant pleural effusion compared with tuberculous pleural effusion. *J Interferon Cytokine Res* 2001;**21**:249−56.
19. Taube JM, Anders RA, Young GD, Xu H, Sharma R, McMiller TL, et al. Colocalization of inflammatory response with B7−h1 expression in human melanocytic lesions supports an adaptive resistance mechanism of immune escape. *Sci Transl Med* 2012;**4**:127ra37.
20. Topalian SL, Drake CG, Pardoll DM. Targeting the PD-1/B7-H1(PD-L1) pathway to activate antitumor immunity. *Curr Opin Immunol* 2012;**24**:207−12.
21. Schneider H, Downey J, Smith A, Zinselmeyer BH, Rush C, Brewer JM, et al. Reversal of the TCR stop signal by CTLA-4. *Science* 2006;**313**:1972−5.
22. Riley JL, Mao M, Kobayashi S, Biery M, Burchard J, Cavet G, et al. Modulation of TCR-induced transcriptional profiles by ligation of CD28, ICOS, and CTLA-4 receptors. *Proc Natl Acad Sci USA* 2002;**99**:11790−5.
23. Qureshi OS, Zheng Y, Nakamura K, Attridge K, Manzotti C, Schmidt EM, et al. Trans-endocytosis of CD80 and CD86: a molecular basis for the cell-extrinsic function of CTLA-4. *Science* 2011;**332**:600−3.
24. Krummel MF, Allison JP. CTLA-4 engagement inhibits IL-2 accumulation and cell cycle progression upon activation of resting T cells. *J Exp Med* 1996;**183**:2533−40.
25. Waterhouse P, Penninger JM, Timms E, Wakeham A, Shahinian A, Lee KP, et al. Lymphoproliferative disorders with early lethality in mice deficient in CTLA-4. *Science* 1995;**270**:985−8.
26. Gabriel EM, Lattime EC. Anti-CTL associated antigen 4: are regulatory T cells a target? *Clin Cancer Res* 2007;**13**:785−8.
27. Leach DR, Krummel MF, Allison JP. Enhancement of antitumor immunity by CTLA-4 blockade. *Science* 1996;**271**:1734−6.
28. van Elsas A, Hurwitz AA, Allison JP. Combination immunotherapy of B16 melanoma using anti-cytotoxic T lymphocyte-associated antigen 4 (CTLA-4) and granulocyte/macrophage colony-stimulating factor (GM-CSF)-producing vaccines induces rejection of subcutaneous and metastatic tumors accompanied by autoimmune depigmentation. *J Exp Med* 1999;**190**:355−66.
29. Phan GQ, Yang JC, Sherry RM, Hwu P, Topalian SL, Schwartzentruber DJ, et al. Cancer regression and autoimmunity induced by cytotoxic T lymphocyte-associated antigen 4 blockade in patients with metastatic melanoma. *Proc Natl Acad Sci USA* 2003;**100**:8372−7.
30. Ribas A, Camacho LH, Lopez-Berestein G, Pavlov D, Bulanhagui CA, Millham R, et al. Antitumor activity in melanoma and anti-self responses in a phase I trial with the anti-cytotoxic T lymphocyte-associated antigen 4 monoclonal antibody CP-675,206. *J Clin Oncol* 2005;**23**:8968−77.
31. Hodi FS, Butler M, Oble DA, Seiden MV, Haluska FG, Kruse A, et al. Immunologic and clinical effects of antibody blockade of cytotoxic T lymphocyte-associated antigen 4 in previously vaccinated cancer patients. *Proc Natl Acad Sci USA* 2008;**105**:3005−10.
32. Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB, et al. Improved survival with ipilimumab in patients with metastatic melanoma. *N Engl J Med* 2010;**363**:711−23.
33. Lynch TJ, Bondarenko I, Luft A, Serwatowski P, Barlesi F, Chacko R, et al. Ipilimumab in combination with paclitaxel and carboplatin as first-line treatment in stage IIIB/IV non-small-cell lung cancer: results from a randomized, double-blind, multicenter phase II study. *J Clin Oncol* 2012;**30**:2046−54.
34. Wolchok JD, Hoos A, O'Day S, Weber JS, Hamid O, Lebbe C, et al. Guidelines for the evaluation of immune therapy activity in solid tumors: immune-related response criteria. *Clin Cancer Res* 2009;**15**:7412−20.
35. Miller AB, Hoogstraten B, Staquet M, Winkler A. Reporting results of cancer treatment. *Cancer* 1981;**47**:207−14.
36. Reuben JM, Lee BN, Li C, Gomez-Navarro J, Bozon VA, Parker CA, et al. Biologic and immunomodulatory events after CTLA-4 blockade with ticilimumab in patients with advanced malignant melanoma. *Cancer* 2006;**106**:2437−44.
37. Pfizer Inc. press release − Pfizer announces discontinuation of phase III clinical trial for patients with advanced melanoma. Available at: http://www.pfizer.com/news/press release/press release detail/pfizer announces

discontinuation of phase III clinical trial for patients with advanced melanoma. [Date accessed: 06 Nov 2015].
38. Calabrò L, Morra A, Fonsatti E, Cutaia O, Amato G, Giannarelli D, et al. Tremelimumab for patients with chemotherapy-resistant advanced malignant mesothelioma: an open-label, single-arm, phase 2 trial. *Lancet Oncol* 2013;**14**:1104−11.
39. Keir ME, Liang SC, Guleria I, Latchman YE, Qipo A, Albacker LA, et al. Tissue expression of PD-L1 mediates peripheral T cell tolerance. *J Exp Med* 2006;**203**:883−95.
40. Dong H, Strome SE, Salomao DR, Tamura H, Hirano F, Flies DB, et al. Tumor-associated B7-H1 promotes T-cell apoptosis: a potential mechanism of immune evasion. *Nat Med* 2002;**8**:793−800.
41. Nishimura H, Nose M, Hiai H, Minato N, Honjo T. Development of lupus-like autoimmune diseases by disruption of the PD-1 gene encoding an ITIM motif-carrying immunoreceptor. *Immunity* 1999;**11**:141−51.
42. Nishimura H, Okazaki T, Tanaka Y, Nakatani K, Hara M, Matsumori A, et al. Autoimmune dilated cardiomyopathy in PD-1 receptor-deficient mice. *Science* 2001;**291**:319−22.
43. Fanoni D, Tavecchio S, Recalcati S, Balice Y, Venegoni L, Fiorani R, et al. New monoclonal antibodies against B-cell antigens: possible new strategies for diagnosis of primary cutaneous B-cell lymphomas. *Immunol Lett* 2011;**134**:157−60.
44. Rizvi NA, Mazieres J, Planchard D, Stinchcombe TE, Dy GK, Antonia SJ, et al. Activity and safety of nivolumab, an anti-PD-1 immune checkpoint inhibitor, for patients with advanced, refractory squamous non-small-cell lung cancer (CheckMate 063): a phase 2, single-arm trial. *Lancet Oncol* 2015;**16**:257−65.
45. Gettinger SN, Horn L, Gandhi L, Spigel DR, Antonia SJ, Rizvi NA, 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**:2004−12.
46. Brahmer J, Reckamp KL, Baas P, Crinò L, Eberhardt WEE, Poddubskaya E, et al. Nivolumab versus docetaxel in advanced squamous-cell non-small-cell lung cancer. *N Engl J Med* 2015;**373**:123−35.
47. Borghaei H, Paz-Ares L, Horn L, Spigel DR, Steins M, Ready NE, et al. Nivolumab versus docetaxel in advanced nonsquamous non-small-cell lung cancer. *New Engl J Med* 2015;**373**:1627−39.
48. Gettinger SN, Hellmann MD, Shepherd FA, Antonia SJ, Brahmer JR, Chow LQM, et al. First-line monotherapy with nivolumab (NIVO; anti-programmed death-1 [PD-1]) in advanced non-small cell lung cancer (NSCLC): safety, efficacy and correlation of outcomes with PD-1 ligand (PD-L1) expression. *J Clin Oncol* 2015;**33**(Suppl):abstract 8025.
49. Antonia SJ, Brahmer JR, Gettinger SN, Chow LQM, Juergens RA, Shepherd FA, et al. Nivolumab (anti-PD-1; BMS-936558, ONO-4538) in combination with platinum-based doublet chemotherapy (PT-DC) in advanced non-small cell lung cancer (NSCLC). *J Clin Oncol* 2014; **32**(Suppl):abstract 8113.
50. Rizvi NA, Chow LQM, Borghaei H, Shen Y, Harbison C, Alaparthy S, et al. Safety and response with nivolumab (anti-PD-1; BMS-936558, ONO-4538) plus erlotinib in patients (pts) with epidermal growth factor receptor mutant (EGFR MT) advanced NSCLC. *J Clin Oncol* 2014; **32**(Suppl):abstract 8022.
51. Antonia SJ, Gettinger SN, Chow LQM, Juergens RA, Borghaei H, Shen Y, et al. Nivolumab (anti-PD-1; BMS-936558, ONO-4538) and ipilimumab in first-line NSCLC: Interim phase I results. *J Clin Oncol* 2014;**32**(Suppl): abstract 8023.
52. Garon EB, Rizvi NA, Hui R, Leighl N, Balmanoukian AS, Eder JP, et al. Pembrolizumab for the treatment of non-small-cell lung cancer. *New Engl J Med* 2015;**372**:2018−28.
53. FDA. Approved drugs. Available at: http://www.fda.gov/Drugs/InformationOnDrugs/ApprovedDrugs/ucm465650.htm. [Date accessed: 06 Nov 2015].
54. Papadimitrakopoulou V, Patnaik A, Borghaei H, Stevenson J, Gandhi L, Gubens MA, et al. Pembrolizumab (pembro; MK-3475) plus platinum doublet chemotherapy (PDC) as front-line therapy for advanced non-small-cell lung cancer (NSCLC): KEYNOTE-021 Cohorts A and C. *J Clin Oncol* 2015;**33**(Suppl):abstract 8031.
55. Patnaik A, Socinski MA, Gubens MA, Gandhi L, Stevenson J, Bachman RD, et al. Phase 1 study of pembrolizumab (pembro; MK-3475) plus ipilimumab (IPI) as second-line therapy for advanced non-small-cell lung cancer (NSCLC): KEYNOTE-021 cohort D. *J Clin Oncol* 2015; **33**(Suppl):abstract 8011.
56. Goldberg SB, Gettinger SN, Mahajan A, Herbst RS, Chiang AC, Tsiouris AJ, et al. Activity and safety of pembrolizumab in patients with metastatic non-small-cell lung cancer with untreated brain metastases. *J Clin Oncol* 2015;**33**(Suppl):abstract 8035.
57. Vansteenkiste J, Fehrenbacher L, Spira AI, Mazieres J, Park K, Smith D, et al. Atezolizumab monotherapy vs docetaxel in 2L/3L non-small-cell lung cancer: primary analyses for efficacy, safety and predictive biomarkers from a randomized phase II study (POPLAR). *European Cancer Congress* 2015. abstract LBA14.
58. Spira AI, Park K, Mazières J, Vansteenkiste JF, Rittmeyer A, Ballinger M, et al. Efficacy, safety and predictive biomarker results from a randomized phase II study comparing MPDL3280A vs docetaxel in 2L/3L NSCLC (POPLAR). *J Clin Oncol* 2015;**33**(Suppl):abstract 8010.
59. Besse B, Johnson M, Jänne PA, Garassino M, Eberhardt WEE, Peters S, et al. Phase II, single-arm trial (BIRCH) of atezolizumab as first-line or subsequent therapy for locally advanced or metastatic PD-L1-selected non-small-cell lung cancer (NSCLC). *European Cancer Congress* 2015. abstract LBA16.
60. Spigel DR, Chaft JE, Gettinger SC, Chao BH, Dirix LY, Schmid P, et al. Clinical activity and safety from a phase II study (FIR) of MPDL3280A (anti-PDL1) in PD-L1−selected patients with non-small-cell lung cancer (NSCLC). *J Clin Oncol* 2015;**33**(Suppl):abstract 8028.
61. Liu SV, Powderly JD, Camidge DR, Ready N, Heist RS, Hodi FS, et al. Safety and efficacy of MPDL3280A (anti-PDL1) in combination with platinum-based doublet chemotherapy in patients with advanced non-small-cell lung cancer (NSCLC). *J Clin Oncol* 2015;**33**(Suppl):abstract 8030.
62. Rizvi NA, Brahmer JR, Ou SHI, Segal NH, Khleif S, Hwu WJ, et al. Safety and clinical activity of MEDI4736, an anti-programmed cell death-ligand 1 (PD-L1) antibody, in patients with non-small-cell lung cancer (NSCLC). *J Clin Oncol* 2015;**33**(Suppl):abstract 8032.
63. Gulley JL, Spigel D, Kelly K, Chandler JC, Rajan A, Hassan R, et al. Avelumab (MSB0010718C), an anti-PD-L1 antibody, in advanced NSCLC patients: a phase 1b, open-label expansion trial in patients progressing after platinum-based chemotherapy. *J Clin Oncol* 2015; **33**(Suppl):abstract 8034.
64. Herbst RS, Soria JC, Kowanetz M, Fine GD, Hamid O, Gordon MS, et al. Predictive correlates of response to the anti-PD-L1 antibody MPDL3280A in cancer patients. *Nature* 2014;**515**:563−7.
65. Kerr KM, Tsao MS, Nicholson AG, Yatabe Y, Wistuba II, Hirsch FR. Programmed death-ligand 1 immunohistochemistry in lung cancer: In what state is this art? *J Thorac Oncol* 2015;**10**:985−9.
66. Lawrence MS, Stojanov P, Polak P, Kryukov GV, Cibulskis K, Sivachenko A, et al. Mutational heterogeneity in cancer and the search for new cancer-associated genes. *Nature* 2013;**499**:214−8.