Immune checkpoint inhibitors in lung cancer: current status and future directions

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Abstract: Recently, the immune checkpoint inhibitors that target programmed death 1 (PD-1)/ programmed death ligand 1 (PD-L1) have made a breakthrough in treating advanced non-small cell lung cancer (NSCLC) with the efficacy of approximately 20%; among which, nivolumab has acquired treatment indications in lung squamous cell carcinoma. The inhibitors targeting cytotoxic T lymphocyte associated antigen 4 (CTLA-4) are also undergoing clinical trials. Researches on immune checkpoint inhibitors have been rapidly implemented in a variety of different types of lung cancer, such as small cell lung cancer (SCLC) and locally advanced NSCLC, and these inhibitors began to be applied in combination with some established treatments, including chemotherapy, targeting therapy and radiotherapy. Undoubtedly, the immune checkpoint inhibitors have become a hot spot in the research and treatment of lung cancer. However, many problems wait to be solved, such as searching for ideal biomarkers, constituting the best criteria for curative effect evaluation, exploring different combination treatment models, and clearly understanding the mechanisms of primary or secondary drug resistance. Along with these problems to be successfully solved, the immune checkpoint inhibitors will have more broad applications in lung cancer therapy.

Keywords: Immune checkpoint inhibitors; lung cancer; efficacy; toxicity

Lung cancer is a malignant tumor with the highest morbidity and mortality in China. According to the National Cancer Center registration report, the incidence and mortality rates of lung cancer in 2011 were 48.3/10 million and 39.3/10 million, respectively (1). With the discovery of driver genes and the application of targeting drugs, patients with advanced lung adenocarcinoma have acquired a significantly prolonged survival time in recent one decade (2,3). However, advances in treating lung squamous carcinoma are still inadequate, the overall 5 years survival rate of patients diagnosed with non-small cell lung cancer (NSCLC) is around 17% (4), and the development of new treatments is in urgent need.

In recent years, one of the breakthroughs in cancer therapy is the application of immune checkpoint inhibitors in clinical trials. The immune response of T cell to the antigen is regulated by the balance between a complex inhibitory signal (also known as the "immune checkpoint") and an activated signal. Cytotoxic T lymphocyte associated antigen 4 (CTLA-4) and programmed death 1 (PD-1), two inhibitory stimulating molecules expressed on T cell surface, act as a "brake" in the immune system (5). Once they interact with their corresponding ligands, the activity of T cell is inhibited, and the immune response to the tumor antigen is attenuated, thereby preventing T cell from attacking tumor (6-8). In addition, tumor cells and

infiltrating lymphocytes can suppress the initial immune response by expressing costimulatory molecules, such as programmed death ligand 1 (PD-L1). The inhibitors that target CTLA-4 and PD-L1/LD-1 can block this interaction, and triggers the immune response to achieve anti-tumor effect (9). At present, the most heavily studied immune checkpoint inhibitors are several antibodies such as anti-CTLA-4 antibody (ipilimumab), anti-PD-1 (nivolumab, pembrolizumab) and anti-PD-L1 antibodies (Mpdl3280a and MEDI-4736), which have shown success in some areas of lung cancer treatment. The relevant clinical and basic researches are still in full swing; here we summarized some of them as follows.

CTLA-4 inhibitor

Ipilimumab is a humanized IgG1 anti-CTLA-4 monoclonal antibody that can bind to CTLA-4, and inhibits the interaction between CTLA-4 and its ligands (CD80/ CD86, also known as B7). The blockade of CTLA-4 signal attenuates the negative regulatory signals that act on T cell, thus enhance the anti-tumor activity of T cell. As opposed to advanced melanoma (10), treatment of lung cancer with ipilimumab alone showed no obvious therapeutic effects (11). While combined with chemotherapy, ipilimumab appeared to have a somewhat clinical benefit in NSCLC and small cell lung cancer (SCLC) treatment (12,13). A multicenter, randomized stage II study was carried out to evaluate the clinical activity of ipilimumab in combination with paclitaxel and carboplatin in the treatment of early and advanced NSCLC. In this study, the patients were randomly divided into ipilimumab plus chemotherapy group and placebo plus chemotherapy group. The ipilimumab plus chemotherapy group had two different modes which are called phased treatment (two cycles of standard chemotherapy, followed by four cycles of ipilimumab combined with standard chemotherapy) and concurrent treatment (four cycles of ipilimumab combined with standard chemotherapy, followed by two cycles of standard chemotherapy); No matter in phased group or concurrent group, if the patient had a stable disease or effective treatment effects during the period of chemotherapy, they continued to receive ipilimumab treatment. A total of 204 cases of NSCLC patients were candidates for the study, and immune-related progression-free survival (irPFS) is the main observation point. In compared to control group, phased treatment significantly improved the patients' irPFS (5.7 $vs.$ 4.6 months, HR =0.82, P=0.05) rather than the overall survival (OS) (12.2 $vs.$ 8.3 months). Interestingly, this irPFS benefit was mainly observed in patients with squamous cell carcinoma rather than non-squamous cell carcinoma, according to the subgroup analysis. On the contrary, the concurrent treatment did not improve patients' irPFS, but shorten the patients' OS (12). Similar studies were also performed in 130 patients with extensive stage SCLC, which showed that patients' irPFS was significantly prolonged in phased treatment group in compared to chemotherapy alone group (6.4 $vs.$ 5.3 months, HR =0.64, P=0.03) (13). Thus, a number of stage III clinical studies on the effect of ipilimumab in lung cancer treatment have been carried out. The global, multicenter, stage III randomized control research [CA184-104] was specifically focused on patients with advanced squamous cell lung carcinoma of the to compare the OS between ipilimumab plus standard chemotherapy and placebo plus standard chemotherapy. CA184-156 and CA184-R62 were the stage III randomized control researches performed in global and Chinese patients. These results will give systematically evaluation on the efficacy, adverse reactions and therapeutic value of ipilimumab in SCLC treatment.

PD-1 inhibitors

Inhibitors of PD-1/PD-L1 relieve the inhibitory state of T cell and promote T cell to attack tumor cells mainly by blocking the interaction between PD-1 receptor and its ligands, such as PD-L1 (B7-H1) and PD-L2 (B7-DC). Nivolumab and pembrolizumab (MK-3475) are two representative PD-1 inhibitors.

Nivolumab is a humanized IgG4 monoclonal antibody that inactivates PD-1 receptors. Stage I dose range expansion column generation study evaluated the efficacy and safety of nivolumab in 296 cases of patients with advanced solid tumors, including 129 cases of advanced NSCLC patients. Fifty-four percent of patients had received more than or equal to three lines of anti-tumor treatment, and patients were randomly divided into three groups to receive three dose levels (1, 3, 10 mg/kg) every 2 weeks, respectively (14). One hundred and twenty-two cases of NSCLC patients can be evaluated on the effects. Among them, 17.1% of patients reached overall response rate (ORR), the 1, 2 and 3 years survival rates were 42%, 24% and 18% respectively, and the OS is 9.9 months (15). Further analysis showed that in all patients, the ORR in the 3 mg/kg dose group was the highest up to 32%, and the OS was the longest for 14.9 months, so the 3 mg/kg was

selected as the standard dose for next studies. The OS of patients with squamous cell carcinoma or with squamous cell carcinoma were 33% and 12%, respectively. But their ORR had no significant difference. The overall incidence rate of drug-related adverse events (AEs) was 41%, grade 3/4 drug-related AEs occurrence rate was 14%, mainly including gastrointestinal tract reaction, pneumonia, skin reactions. The total incidence rate of drug associated pneumonia is 3%, grade 3/4 pneumonia occurrence rate is 1%, 3 patients died of pulmonary toxicity (2 cases of NSCLC and 1 cases colon). The adverse reactions of patients in lung cancer group and the whole group were similar (14). A long-term observation on the drug safety showed that 3 patients (2%) in the lung cancer group died of the drug related pneumonia (15). PD-L1 expression in tumor cells of 42 patients samples were examined via immunohistochemical staining, the ORR of PD-L1 positive patients is significantly higher than that of PD-L1 negative patients (36% *vs.* 0%, P=0.006), implicating that the expression of PD-L1 in tumor cells correlates with ORR (14). Then the global multicenter, single arm stage II clinical research (CheckMate 063) evaluated the effect of nivolumab in the patients with advanced lung squamous carcinoma, including 117 patients who had received second-line chemotherapy at least , the ORR was 14.5%, the median to onset time was 3.3 months, and the median duration of efficacy was 6 months; 17% of patients occurred treatment related toxicities of grade 3/4 , including fatigue, pneumonia and diarrhea; two cases occurred therapy related death, one case died of pneumonia (16). A number of clinical studies on the effect of nivolumab in lung cancer treatment were reported at the 2015 ASCO meeting. The stage III randomized control study recruited 272 patients, and compared the effect between nivolumab and docetaxel in the second and third line therapy, the OS of nivolumab group was better that that of docetaxel group ((9.2 *vs.* 6.5 months, HR =0.59, P=0.00025), both PFS (3.5 *vs.* 2.8 months, HR =0.62, P=0.0004) and the ORR (20% *vs.* 9%, P=0.0083) increased strikingly, but the PD-L1 expression did not correlate with curative effect and prognosis. However, it is gratified that the drug related grade 3/4 AEs incidence of nivolumab was only 7%, and there was no treatment related death (17). CheckMate-057 studies were performed in the NSCLC, and all the ORR, PFS and OS in the nivolumab group were significantly better than that in the docetaxel group. Japanese scholars reported the effect of nivolumab in the treatment of stage II Asian patients. In the 111 cases of patients with advanced

recurrence, the ORR of squamous cell carcinoma patients and non-squamous cell carcinoma patients were 25.7% and 19.7%, respectively (18). They also reported a stage I study on the effect of nivolumab in the first-line treatment of advanced NSCLC, which includes 52 cases of squamous patients and NSCLC patients, the ORR was 21%, the median OS was 98.3 weeks, the ORR of PD-L1 positive patients was higher than that of negative patients (31% *vs.* 10%), but there was no difference between the OS of two groups (19). In a stage I/II SCLC study (CheckMate 032), 86 cases with relapsed SCLC were treated with nivolumab plus or not plus ipilimumab therapy, the ORR of combined drug group and single drug group was 18.0% and 17.4%, respectively, PD-L1 expression is independent with curative effect, and the incidence of AEs was higher in the combined drug group (20). So far, the therapeutic effect of nivolumab in the treatment of advanced NSCLC (including squamous and non-squamous cell carcinoma) has been fully affirmed, and nivolumab shows excellent promise in the first-line treatment of advanced NSCLC and the save treatment of relapsed SCLC. FDA approved the application of nivolumab in the treatment of metastatic or late stage NSCLC in March 2015. Further research and exploration are still continuing, such as whether first-line treatment is better that chemotherapy, whether nivolumab can be used in combination with other therapies and therapeutic drugs, and how to combine two kinds of immune check point inhibitors.

Pembrolizumab (MK-3475) is another kind of humanized monoclonal antibody of IgG4-κ type, which blocks PD-1 function with high selectivity through the similar mechanism as nivolumab. The stage I study (KEYNOTE-001) was performed in 495 cases of candidates with locally advanced/advanced NSCLC, including 101 initial treated patients and 394 treated patients. Pembrolizumab was used by 2 mg/kg every 3 weeks, or 10 mg/kg every 2 weeks, and the patients were divided into study group of 182 cases and verify group of 313 cases, the PD-L1 expression in tumor cells was determined by immunohistochemical staining. The results showed that the most common adverse reactions were fatigue, skin itching and decreased appetite, the incidence rate of AEs of more than or equal to 3 degrees was 9.5%, among which the incidence rate of pneumonia was 1.8%, 1 cases of pneumonia (0.2%) patients died; the ORR was 19.4%, the ORR of initial treatment and retreatment patients were 24.8% and 18.0%, and the median response duration was 12.5 months, the mFPS was 3.7 months, the

median OS was 12.0 months (21). The ORR of PD-L1 positive patients was 45.2%, the mPFS was 6.3 months and the median survival time was not achieved, all these were obviously better that that of PD-L1 negative patients. Pembrolizumab showed a strong anti-tumor activity in advanced NSCLC, adverse reactions were tolerated, and PD-L1 (+) could predict the therapeutic efficacy. A series of exploratory studies on pembrolizumab were also reported at the ASCO conference in 2015. Pembrolizumab was applied in combination with platinum in the first-line treatment of stage IIIB/IV NSCLC (KEYNOTE-021 Cohort A and group C), a total of 44 patients were tested, and the ORR of pembrolizumab combined with taxol plus carboplatin or pemetrexed plus carboplatin was 30% and 58%, respectively. The drug safety was in reasonable and controllable range (22). The KEYNOTE-021 Cohort D study assessed the combination effect of two kinds of immune targeting drugs, pembrolizumab was combined with ipilimumab in the treatment of advanced and recurrent NSCLC, in the 11 cases which can be evaluated the efficacy, 1 case was completely remission (CR) (9%), 5 cases were partial remission (PR) (45%), all patients had stable diseases for more than or equal to 6 weeks, this combination therapy displayed a strong anti-tumor activity, and the toxicity can be tolerated (23). Another study was performed in a small sample of patients with brain metastases from asymptomatic, and untreated NSCLC, the ORR of 10 patients with brain lesions was 44% after pembrolizumab treatment (24). Ott *et al.* reported the KEYNOTE-028 research, in which 20 cases of patients who had PD-L1 (+) and extensive stage SCLC and had failures in chemotherapy were received pembrolizumab treatment, the ORR was 35%, the effective treatment duration was more than 16 weeks (25). These small sample studies lay a good foundation for the expansion of pembrolizumab in the treatment of indications. Currently, several large clinical studies are being carried out: the stage II/III clinical research KEYNOTE-010 which randomly compares the effect of pembrolizumab and docetaxel in the second-line treatment of advanced NSCLC; the stage III randomized control study (KEYNOTE-024, KEYNOTE-042) which compares the effects between pembrolizumab and platinum doublet chemotherapy in the first-line treatment of PD-L1 positive metastatic/advanced NSCLC patients.

PD-L1 inhibitors

MPDL3280A is a human IgG4 antibody against PD-L1. In stage I research, 277 cases of patients with various advanced tumors received the MPDL3280A single drug treatment for 1 time every 3 weeks. The results showed that the AEs level of grade 3/4 was 13%, mainly including fatigue, diarrhea, high blood glucose, hypoxia, and higher liver enzymes; 3–5 degree of pneumonia was not observed. Compared with the PD-1 inhibitor, the tolerance of MPDL3280A appeared better (26). A total of 175 patients could be evaluated for efficacy, the ORR was 18%, and the mPFS was 18 weeks. Among them, the efficacy of MPDL3280A on NSCLC is 23%, and the efficiency rate of smoking subgroup was significantly higher than that of non-smokers (42% *vs.* 10%). The PD-L1 expression of tumor infiltrating lymphocytes (infiltrating lymphocytes Tumor, TILs) was detected in the pre-treatment samples, which showed that in patients with NSCLC, the ORR of patients with PD-L1 expression (3+) or patients with no PD-L1 expression was 83% and 20% (P=0.015), respectively. In the whole group of patients, the ORR of patients with PD-L1 expression (3+) or patients with no PD-L1 expression was 46% and 13% (P=0.0007), respectively. The PFS was 37.3 and 8.1 months, respectively (26). At the 2015 ASCO conference, a randomized control study of stage II compared the effect of MPDL3280A with docetaxel in the second or third line treatment for advanced NSCLC (POPLAR study). In this study, 287 patients were enrolled, MPDL3280A could significantly improve the ORR, PFS, OS and TILs of patients in compared with chemotherapy. Patients with PD-L1 expression (+) in tumor cells could benefit more from the treatment (27). In another stage II study, MPDL3280A was used to treat IIIB/IV phase NSCLC with PD-L1 expression (+) (FIR study). The patients were divided into three treatment groups, the first group was patients with initial treatment; the second group was patients with no cerebral metastasis in the second line or above; the third group was patients with no symptoms of cerebral metastasis in the second line or above. A total of 138 patients were enrolled and 114 cases could be evaluated for the efficacy. The incidence of treatment-associated AEs level of grade 3/4 was 15%; both patients with initial treatment and treated patients had a good curative effect; patients with PD-L1 expression (3+) had a higher ORR (28). Horn *et al.* reported that in 88 cases of recurrent NSCLC patients, the ORR was significantly higher in patients with PD-L1 expression (3+) than in other patients (45% *vs.* 14%) (29). According to the MPDL3280A related researches, the positive expression of PD-L1 is a good index for predicting the remission rate, the effective rate of patients with high expression (3+) can be as

high as 45–83% (27-29). MPDL3280A related follow-up studies were almost performed in PD-L1 positive patients, currently a number of II, III stage of clinical trial are still undergoing.

MEDI-4736 is another IgG1-k type of PD-L1 inhibitor, which blocks the binding between PD-L1 and PD-1 or CD80. Stage I studies on the treatment effect of MEDI-4736 in multiple types of cancers were reported at the 2014 ASCO conference. A total of 346 patients with advanced cancer were enrolled in the clinical study, including 143 patients with NSCLC, MEDI-4736 showed curative effect on many tumors (30). At the 2015 ASCO conference, it was reported that 198 cases of patients with NSCLC, 116 cases of patients with non-squamous cell carcinoma and 82 cases of patients with squamous cell carcinoma were treated with MEDI-4736. MEDI-4736 was applied at 10 mg/kg, 1 time per 2 weeks. The incidence of drug related AEs was 48%, mainly including fatigue (14%), loss of appetite (9%), and nausea (8%). The incidence of pneumonia was 1%, both 1–2 degrees and the incidence of AEs more than 3 degrees was 6%. One hundred and forty-nine cases could be evaluated for efficacy, the ORR was 14% and the effect was sustained. The ORR of PD-L1 positive patients was 23%, The ORR of patients with squamous cell carcinoma were higher than those of non-squamous cell carcinoma (21% *vs.* 10%) (31). Other PD-L1 inhibitors, such as avelumab (MSB0010718C) have also cut a striking figure in the treatment of NSCLC (32). These results suggest that PD-L1 inhibitors are effective in the treatment of advanced NSCLC, the drug tolerance is good and PD-L1 (+) patients tend to have better therapeutic effects.

Seek biomarkers for predicting efficacy

The efficacy of PD-1 and PD-L1 inhibitors in the treatment of advanced NSCLC was approximately 20%, there are still about 80% of patients resistant to the treatment, therefore, and seeking biomarkers for predicting efficacy is in urgent need. PD-L1 is currently the most widely studied biological marker, it was reported that PD-L1 expressed in a variety of tumors and about 27–50% of lung cancer patients showed PD-L1 positive expression (33,34). Is PD-L1 a potential ideal predictor? According to the present researches, the situation is quite complex. Firstly, the expression of PD-L1 is not limited to cancer cells, but also in tumor associated macrophages, dendritic cells, fibroblasts and activated T cells (35). Besides, PD-L1 is a dynamic biomarker, as its expression will fluctuate during the whole disease process;

the expression of PD-L1 is variable with time, or can be induced by past treatment and other factors. Secondly: many studies showed that PD-L1 (+) can predict the short-term efficacy of PD-1 or PD-L1 like drugs (14,26-29,31,36), but only a few studies supported that it can predict survival benefit (26,36). Thirdly, this kind of drugs is also effective in patients with PD-L1 expression (–) (16,37). Fourthly, the predictive value of PD-L1 is not consistent with the various PD-1/PD-L1 like inhibitors (16,26-29,31,37). Fifthly, there are still many problems in the detection of PD-L1. On one hand, the methods and the antibodies used to analyze the expression of PD-L1 were not unified in different clinical researches. On the other hand, there are differences in the observed objects, some of researchers analyzed the expression of PD-L1 in tumor cells (21,36), and some of researches also detected the PD-L1 expression in infiltrating lymphocytes (26,27). Finally, the definitions of cut-off values of PD-L1 positive are not uniform in different studies. Taken together, it is not difficult to find that detection of PD-L1 expression is still not mature, and several different detection techniques and analysis methods are still in development. Therefore, it is urgent to establish the standardization of PD-L1 testing technology and interpretation standards. The future research directions are searching for other potential biological markers that predicting efficacy, such as CD8^{+} expressed in T cells or cytokines expressed in tumor samples (38). In a word, the expression of PD-L1 alone cannot accurately evaluate the dynamic immune microenvironment; it is also unable to predict all possible effective patients according to the cut-off values of PD-L1. Therefore, it is difficult to find ideal biomarkers for predicting efficacy. Even so, it is still able to screen out more effective patients by detecting the expression of PD-L1. So, PD-L1 remains the most promising biomarker for PD-1/PD-L1 like drugs at this stage.

Drug for which target is better?

CTLA-4, PD-1 and PD-L1, CTLA-4 and PD-1, all of which belongs to inhibitory receptors, are the main targets of the current immune targeted therapy. The formers are expressed in the T cells involved in initial antigen reaction (activated CD8^{+} T cells), the latter genes are expressed in activated T cells, B cells, NK cells, and different types of tumor infiltrating lymphocytes. The efficacy of anti-PD-1 antibody was better than that of anti-CTLA-4 antibody in the treatment of melanoma (39). In the study of NSCLC,

treatment with the CTLA-4 antibodies alone had little effect, and single drugs that blocks PD-1/PD-L1 all showed tumor activities.

PD-1 has two ligands, PD-L1 and PD-L2; PD-L1 has two receptors, PD-1 and CD80 (B7.1). Although anti PD-1 antibodies and anti PD-L1 antibodies are all targeted for the PD-1/PD-L1 signaling axis, blocking PD-1 is not equivalent to blocking PD-L1. Anti-PD-1 antibodies could block the binding between PD1 and PD-L1 or PD-L2, but not the interaction between PD-L1 and CD80. Accordingly, the anti-PD-L1 antibodies could block the binding between PD-L1 and PD-1 or CD80, but not the interaction between PD-1 and PD-L2 (40). At present, PD-L1 antibodies seem to be more dominant in adverse reactions from clinical research. The combination of PD-1 and anti PD-L1 antibodies are found more efficient in preclinical models (41). Therefore, whether there are differences in the clinical efficacy and toxicity between PD-1 and anti PD-L1 antibodies, and whether the combination of the two kind antibodies has more practical significance is need to be confirmed by clinical trials.

The most appropriate treatment group

Ipilimumab appears a better effect on patients with squamous cell carcinoma (13,14,17), and nivolumab is currently used in the treatment of NSCLC. Interestingly, some studies have found that smoking is also a factor affecting the efficacy of PD-1/PD-L1 antibodies (26,42). The vast majority of NSCLC have a long history of smoking, whether smoking leads to better immune response to lung cancer and how to explain this phenomenon? At present, some studies demonstrated that gene mutations are more complex in smoking patient, and the more the numbers of gene mutations, the more likely immune cells are able to recognize tumor cells, and the immune therapy may be more efficient (43-45). However, this theory could not explain all the phenomena; for example, there were no differences in efficacy of some immune checkpoint inhibitors in different pathological subtypes; the immune checkpoint inhibitors are effective in only a few patients, even in patients with squamous cell carcinoma. Therefore, it is worth to further exploring how to select the most appropriate treatment group according to pathological subtype, molecular markers, clinical and genetic features.

Correlation between the drive gene and PD-L1

Drive genes have been identified in about 64% of lung adenocarcinoma (46). For the correlation between EGFR mutation and PD-L1 expression, different conclusions were obtained. Akbay et al. (47) has reported that the activation of EGFR signaling pathway was associated with the high expression of PD-L1, PD-1 and CTLA-4 in the EGFR-driven mouse lung cancer and human NSCLC cell line (48-50). In contrast, other researchers have shown that there was no significant correlation between the expression of PD-L1 and drive genes (51). There are also studies showing that the expression level of PD-L1 was dynamically changed before and after treatment in the patients with EGFR mutation and ALK (+) (52). Further studies on basic research have revealed that EGFR could raise the expression of PD-L1 through activating the p-ERK1/2/p-c-Jun signaling pathway; EGFR TKI could not only directly inhibit the activity of tumor cells, but also indirectly enhance the anti-tumor immunity of body by suppressing the expression of PD-L1. So, the researchers considered that development of PD-1/PD-L1 antibodies could be an important therapeutic direction for EGFR-TKI mutation sensitive patients, especially for drug resistance patients (53). Meanwhile, there are also researches focused on the correlation between the mutated genes like K-RAS and expression of PD-L1 (49). Recent studies have reported that the genomic landscape of lung cancer would affect the efficacy of PD-1 antibodies (54). Therefore, it is very important to investigate how to combine the target drug with immune checkpoint inhibitors for clinical treatment.

Summary

Immune checkpoint inhibitors in lung cancer therapy have shown promise. Nivolumab has preempted in second-line therapy of lung squamous cell carcinoma, similar drugs and related research are undergoing, but there are still many unsolved problems: (I) searching for biomarkers of ideal prediction effect; (II) determining the best curative effect evaluation criteria; (III) deep understanding primary and secondary resistance mechanism; (IV) investigating different modes of treatment: first-line, second-line or third line treatment? Single or combined therapy? Combined chemotherapy, radiotherapy or targeted drug (55)? The combination of two kinds of immune checkpoint inhibitors has been confirmed in the treatment of advanced melanoma (56), whether it has the same effect in patients with lung cancer? With these problems to be solved, it is believed that the immune checkpoint inhibitors will revolutionize the clinical practice of lung cancer.

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Footnote

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References

1. Chen W, Zheng R, Zeng H, et al. Annual report on status of cancer in China, 2011. Chin J Cancer Res 2015;27:2-12.

2. Jänne PA, Engelman JA, Johnson BE. Epidermal growth factor receptor mutations in non-small-cell lung cancer: implications for treatment and tumor biology. J Clin Oncol 2005;23:3227-34.

3. Kwak EL, Bang YJ, Camidge DR, et al. Anaplastic lymphoma kinase inhibition in non-small-cell lung cancer. N Engl J Med 2010;363:1693-703.

4. Molina JR, Yang P, Cassivi SD, et al. Non-small cell lung cancer: epidemiology, risk factors, treatment, and survivorship. Mayo Clin Proc 2008;83:584-94.

5. Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. Nat Rev Cancer 2012;12:252-64.

6. Greenwald RJ, Freeman GJ, Sharpe AH. The B7 family revisited. Annu Rev Immunol 2005;23:515-48.

7. Latchman Y, Wood CR, Chernova T, et al. PD-L2 is a second ligand for PD-1 and inhibits T cell activation. Nat Immunol 2001;2:261–8.

8. Fife BT, Bluestone JA. Control of peripheral T-cell tolerance and autoimmunity via the CTLA-4 and PD-1 pathways. Immunol Rev 2008;224:166-82.

9. Brahmer JR. Harnessing the immune system for the treatment of non small-cell lung cancer. J Clin Oncol 2013;31:1021-8.

10. Hodi FS, O'Day SJ, McDermott DF, et al. Improved survival with ipilimumab in patients with metastatic melanoma. N Engl J Med 2010;363:711-23.

11. Zatloukal P, Heo DS, Park K, et al. Randomized phase II clinical trial comparing tremelimumab (CP-675, 206) with best supportive care (BSC) following first-line platinum-based therapy in patients (pts) with advanced non-small cell lung cancer (NSCLC). J Clin Oncol 2009;27:abstr 8071.

12. Lynch TJ, Bondarenko I, Luft A, 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.

13. Reck M, Bondarenko I, Luft A, et al. Ipilimumab in combination with paclitaxel and carboplatin as first line therapy in extensive-disease-small-cell lung cancer: results from a randomized, double-blind, multicenter phase 2 trial. Ann Oncol 2013;24:75-83.

14. Topalian SL, Hodi FS, Brahmer JR, et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. N Engl J Med 2012;366:2443-54.

15. Gettinger SN, Horn L, Gandhi L, et al. Overall Survival and Long-Term Safety of Nivolumab (Anti-Programmed Death 1 Antibody, BMS-936558, ONO-4538) in Patients With Previously Treated Advanced Non-Small-Cell Lung Cancer. J Clin Oncol 2015;33:2004-12.

16. Rizvi NA, Mazières J, Planchard D, 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.

17. Spigel DR., Reckamp L, Rizvi NA, et al. A phase III study (CheckMate 017) of nivolumab (NIVO; anti-programmed death-1 (PD-1)) vs docetaxel (DOC) in previously treated advanced or metastatic squamous (SQ) cell non-small cell lung cancer (NSCLC). J Clin Oncol 2015;33:abstr 8009.

18. Nishio M, Hida T, Nakagawa K, et al. Phase II studies of nivolumab (anti-PD-1, BMS-936558, ONO-4538) in patients with advanced squamous (sq) or nonsquamous (non-sq) non-small cell lung cancer (NSCLC). J Clin Oncol 2015;33:abstr 8027.

19. Gettinger N, Hellmann MD, Shepherd FA, 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:abstr 8025.

20. Antonia SJ, Bendell JC, Taylor MH, et al. Phase I/II study of nivolumab with or without ipilimumab for treatment of recurrent small cell lung cancer (SCLC): CA209-032. J Clin Oncol 2015;33:abstr 7503.

21. Garon EB, Rizvi NA, Hui R, et al. Pembrolizumab for the treatment of non-small-cell lung cancer. N Engl J Med 2015;372:2018-28.

22. Papadimitrakopoulou V, Patnaik A, Borghaei H, 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:abstr 8031.

23. Patnaik A, Socinski MA, Gubens MA, 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:abstr 8011.

24. Goldberg SB, Gettinger SN., Mahajan A, 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:abstr 8035.

25. Ott PA, Fernandez ME, Hiret S, et al. Pembrolizumab (MK-3475) in patients (pts) with extensive-stage small cell lung cancer (SCLC): Preliminary safety and efficacy results from KEYNOTE-028. J Clin Oncol 2015;33:abstr 7502.

26. Herbst RS, Soria JC, Kowanetz M, et al. Predictive correlates of response to the anti-PD-L1 antibody MPDL3280A in cancer patients. Nature 2014;515:563-7.

27. Spira A, Park K, Mazières J, 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:abstr 8010.

28. Spigel DR, Chaft JE., Gettinger SN, 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:abstr 8028.

29. Horn L, Spigel DR, Gettinger SN, et al. Clinical activity, safety and predictive biomarkers of the engineered antibody MPDL3280A (anti-PDL1) in non-small cell lung cancer (NSCLC): update from a phase Ia study. J Clin Oncol 2015;33:abstr 8029.

30. Segal NH, Antonia SJ, Brahmer JR, et al. Preliminary data from a multi-arm expansion study of MEDI4736, an anti-PD-L1 antibody. J Clin Oncol 2014;32:abstr 3002.

31. Rizvi NA, Brahmer R, Ou SHI, 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:abstr 8032..

32. Gulley JL, Spigel D, Kelly K, 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:abstr 8034.

33. Konishi J, Yamazaki K, Azuma M, et al. B7-H1 expression on non-small cell lung cancer cells and its relationship with tumor-infiltrating lymphocytes and their PD-1 expression. Clin Cancer Res 2004;10:5094-100.

34. Velcheti V, Schalper KA, Carvajal DE, et al. Programmed death ligand-1 expression in non-small cell lung cancer. Lab Invest 2014;94:107-16.

35. Chen DS, Irving BA, Hodi FS. Molecular pathways: next-generation immunotherapy–inhibiting programmed death-ligand 1 and programmed death-1. Clin Cancer Res 2012;18:6580-7.

36. Rizvi NA, Garon EB, Leighl N, et al. Optimizing PD-L1 as a biomarker of response with pembrolizumab (pembro; MK-3475) as first-line therapy for PD-L1–positive metastatic non-small cell lung cancer (NSCLC): Updated data from KEYNOTE-001. J Clin Oncol 2015;33:abstr 8026.

37. Paz-Ares L, Horn L, Borghaei H, et al. Phase III, randomized trial (CheckMate 057) of nivolumab (NIVO) versus docetaxel (DOC) in advanced non-squamous cell (non-SQ) non-small cell lung cancer (NSCLC). J Clin Oncol 2015;33:abstr LBA109.

38. Tumeh PC, Harview CL, Yearley JH, et al. PD-1 blockade induces responses by inhibiting adaptive immune resistance. Nature 2014;515:568-71.

39. Robert C, Schachter J, Long GV, et al. Pembrolizumab versus Ipilimumab in Advanced Melanoma. N Engl J Med 2015;372:2521-32.

40. Keir ME, Butte MJ, Freeman GJ, et al. PD-1 and its ligands in tolerance and immunity. Annu Rev Immunol 2008;26:677-704.

41. Curran MA, Montalvo W, Yagita H, et al. PD-1 and CTLA-4 combination blockade expands infiltrating T cells and reduces regulatory T and myeloid cells within B16 melanoma tumors. Proc Natl Acad Sci U S A 2010;107:4275-80.

42. Rizvi NA, Hellmann MD, Snyder A, et al.Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer. Science 2015;348:124-8.

43. Melero I, Lasarte JJ. Genetic basis for clinical response to CTLA-4 blockade. N Engl J Med 2015;372:783.

44. Yadav M, Jhunjhunwala S, Phung QT, et al. Predicting immunogenic tumour mutations by combining mass spectrometry and exome sequencing. Nature 2014;515:572-6.

45. Gubin MM, Zhang X, Schuster H, et al. Checkpoint blockade cancer immunotherapy targets tumour-specific mutant antigens. Nature 2014;515:577-81.

46. Kris MG, Johnson BE, Berry LD, et al. Using multiplexed assays of oncogenic drivers in lung cancers to select targeted drugs. JAMA 2014;311:1998-2006.

47. Akbay EA, Koyama S, Carretero J, et al. Activation of the PD-1 pathway contributes to immune escape in EGFR-driven lung tumors. Cancer Discov 2013;3:1355-63.

48. Azuma K, Ota K, Kawahara A, et al. Association of PD-L1 overexpression with activating EGFR mutations in

surgically resected nonsmall-cell lung cancer. Ann Oncol 2014;25:1935-40.

49. D'Incecco A, Andreozzi M, Ludovini V, et al. PD-1 and PD-L1 expression in molecularly selected non-small-cell lung cancer patients. Br J Cancer 2015;112:95-102.

50. Tang Y, Fang W, Zhang Y, et al. The association between PD-L1 and EGFR status and the prognostic value of PD-L1 in advanced non-small cell lung cancer patients treated with EGFR-TKIs. Oncotarget 2015;6:14209-19.

51. Yang CY, Lin MW, Chang YL, et al. Programmed cell death-ligand 1 expression in surgically resected stage I pulmonary adenocarcinoma and its correlation with driver mutations and clinical outcomes. Eur J Cancer 2014;50:1361-9.

52. Gainor JF, Sequist LV, Shaw AT, et al. Clinical correlation and frequency of programmed death ligand-1 (PD-L1) expression in EGFR-mutant and ALK-rearranged non-small cell lung cancer (NSCLC). J Clin Oncol 2015;33:abstr 8012.

53. Chen N, Fang W, Zhan J, et al. Up-regulation of PD-L1 by EGFR Activation Mediates the Immune Escape in EGFR-driven NSCLC: Implication forOptional Immune Targeted Therapy for NSCLC Patients with EGFR Mutation. J Thorac Oncol 2015;10:910-23.

54. Rizvi NA, Hellmann MD, Snyder A, et al. Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer. Science 2015;348:124-8.

55. Twyman-Saint Victor C, Rech AJ, Maity A, et al. Radiation and dual checkpoint blockade activate non-redundant immune mechanisms in cancer. Nature 2015;520:373-7.

56. Postow MA, Chesney J, Pavlick AC, et al. Nivolumab and Ipilimumab versus Ipilimumab in Untreated Melanoma. N Engl J Med 2015;372:2006-17.