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REVIEW

Review and perspective on adjuvant and neoadjuvant immunotherapies in NSCLC

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Abstract: Postoperative patients have risk recurring, even for completed resected early stage non-small-cell lung cancer (NSCLC). To control the recurrence rate, neoadjuvant and adjuvant therapies have been applied widely in clinical practice; however, neoadjuvant and adjuvant immunotherapy clinical trials on NSCLC are still being explored. In this review, we summarized the research progress and outline the issues need to be solved on adjuvant and neoadjuvant immunotherapies in NSCLC.

Keywords: adjuvant immunotherapies, neoadjuvant immunotherapies, immune checkpoint inhibitor, non-small-cell lung cancer, biomarker, liquid biopsy

Introduction

The data of global cancer statistics 2018 showed that lung cancer was the most commonly diagnosed cancer (11.6% of all cases) and the leading cause of cancer death (18.4% of the total cancer deaths).[1] Even for postoperative patients with early-stage lung cancer, the rate of death or recurring varied from 8% to 66%.[2,3] Owing to the presence of micro-metastases before surgery, it was tough to control relapses in surgery patients.[4]

To improve prognosis, adjuvant and neoadjuvant chemotherapies are proposed and two meta-analyses of randomized trials testified the survival advantage of adjuvant and neoadjuvant chemotherapies.[5,6] Moreover, immunotherapy is a hot-spot in the treatment of lung cancer. Programmed death-1 (PD-1)/programmed death-ligand 1 (PD-L1) monoclonal antibodies have shown promising efficacy in advanced nonsquamous (p=0.002) and squamous (p<0.001) non-small-cell lung cancer (NSCLC).[7,8] However, adjuvant and neoadjuvant immunotherapies in lung cancer are still worth to be explored. Here, by reviewing the research progress about adjuvant and neoadjuvant immunotherapies (as Figure 1 shown), we summarized these researches and outline the issues need to be solved on adjuvant and neoadjuvant immunotherapies in NSCLC.

Adjuvant immunotherapies

Adjuvant therapies are aimed to improve prognosis and survival for patients with resected NSCLC. Immunotherapies for NSCLC have developed rapidly in recent years. Adjuvant immunotherapies have attracted the researches' attention, and strategies of adjuvant therapies are increasingly diverse. Here, we concluded the studies on adjuvant immunotherapies.

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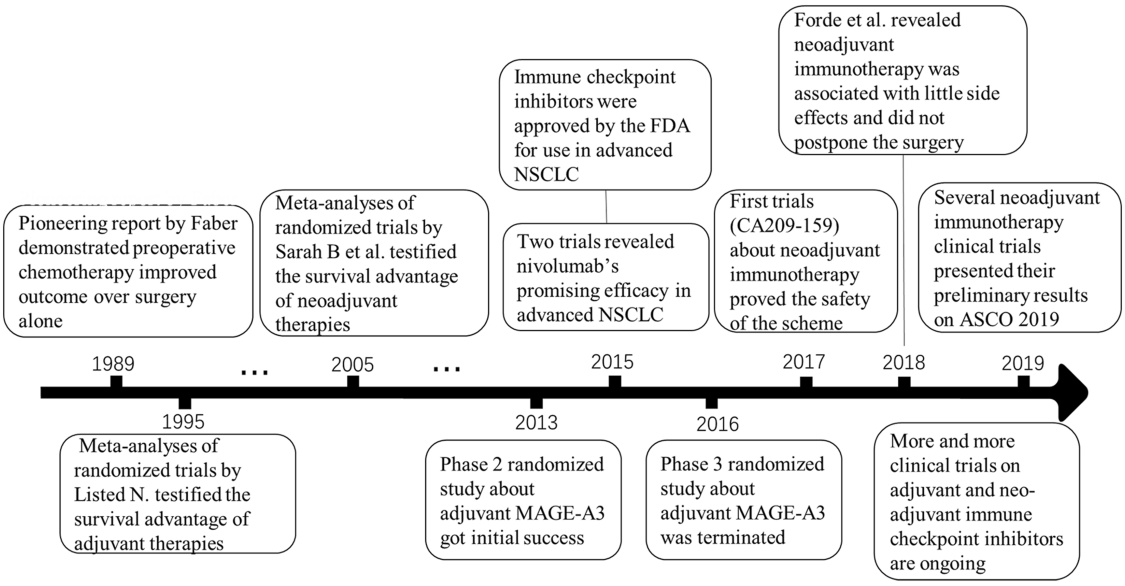

Figure 1 Time axis flowchart on research progress about adjuvant and neoadjuvant immunotherapies.

1. Adjuvant passive immunotherapy

Adjuvant passive immunotherapies have initially focused on the dendritic cell-cytokine induced killer (DC-CIK) and tumor vaccines. A study including 157 patients with stage III NSCLC showed that the median survival time of the patients in the control and adjuvant DC-CIK cell immunotherapy group was 22 months (95% CI, 16.23–27.77) and 28 months (95% CI, 24.39–31.61).[9]

The Melanoma-associated antigen 3 (MAGE-A3) gene was presented to specific T cells by human leukocyte antigen (HLA) molecules at the cell surface as a tumor-specific antigen.[10] MAGE-A3 antigen was a particular interest target for a vaccination strategy. In a double-blind, randomized, placebo-controlled phase II postoperative study,[11] MAGE-A3 immunization did not show significant improvement in disease-free survival (DFS), but the toxicity is controllable. A parallel-group phase I study[12] showed that adjuvant MAGE-A3 could induce MAGEA3-specific immune responses no matter with concurrent chemotherapy or not. In a randomized, double-blind, placebo-controlled trial,[13] adjuvant treatment with the MAGE-A3 immunotherapeutic did not significantly increase DFS compared with placebo in patients with MAGE-A3-positive surgically resected NSCLC and median DFS was 60.5 months for the MAGE-A3 immunotherapeutic group and 57.9 months for the placebo group. These disappointing results led to the discontinuation of further clinical development of the MAGE-A3 immunotherapies.

2. Adjuvant immune checkpoint inhibitors

Immune checkpoint inhibitors, such as PD-1/PD-L1 monoclonal antibodies have been successfully used in advanced lung cancer patients. Immune checkpoint inhibitors anti-PD-1 and PD-L1 antibodies alone,[14] or combined with chemotherapy[15] showed significant overall survival (OS) advantage in stage IV lung cancer. As to the resectable patients, a meta-analyze showed patients might get benefits from adjuvant checkpoint inhibitors (PD-1/PD-L1 inhibitor).[16] Given these positive trials, immune checkpoint inhibitors have been used as adjuvant treatment in some on-going clinical trials, including pembrolizumab (NCT02504372), durvalumab (NCT02273375), atezolizumab (NCT02486718), nivolumab (NCT02595944) (as Table 1 shown). Nevertheless, there hasn't been a standard formulation for adjuvant immune checkpoint inhibitors, neither dosage nor circles of treatment.

Neoadjuvant immunotherapies

1. Advantage of immunotherapies in neoadjuvant strategy

Preoperative chemotherapies combined with surgery had better survival than surgery only.[17,18] However, neoadjuvant therapies didn't show significant longer survival in all studies.[19,20] For immunotherapies, preclinical work suggests that neoadjuvant application of checkpoint inhibitors could be superior to neoadjuvant

Table 1 Clinical trials of adjuvant immunotherapies for NSCLC

Study name	Drug	Sample size	Hazard ratio for OS	Hazard ratio for PFS	Identifier
MAGRIT	GSK1572932A Antigen-Specific Cancer Immunotherapeutic	2312	None	1.02 (95% CI: 0.89–1.18)	NCT00480025
PEARLS	Pembrolizumab	1080 (Estimated)	Ongoing	Ongoing	NCT02504372
BR31	Durvalumab	1360 (Estimated)	Ongoing	Ongoing	NCT02273375
IMpower010	Atezolizumab	1280 (Estimated)	Ongoing	Ongoing	NCT02486718
ANVIL	Nivolumab	903 (Estimated)	Ongoing	Ongoing	NCT02595944

Abbreviations: OS, overall survival; PFS, progression-free survival.

chemotherapy.[21] A clinical trial included 20 patients (adjuvant 10: neoadjuvant 10) with stage III melanoma showed that the rate of death was lower in the neoadjuvant group than that in the adjuvant group.[22]

It has been considered that administration of checkpoint inhibitors before resection maybe induce a stronger and more prolonged antitumor T cell immune response compared to administration of checkpoint inhibitors after surgery, resulting in more effective prevention of tumor relapse.[23] Moreover, massive structure of lymphatic system around lung cancer before resection was relatively intact and checkpoint inhibitors could work better.[24] Also, a hypothesis that higher tumor burden can assist checkpoint inhibitors to stimulate antitumor T cell immune response better before an operation is considerable.

2. Pathological response

Pathological complete response (PCR), defined as eradication of all tumors from resected lung and lymph node tissue, was regarded as a surrogate for OS in neoadjuvant research. Depierre et al[25] investigated 179 patients with stage IB–IIIA NSCLCs treated with neoadjuvant chemotherapy and shown that 11% of patients got a PCR and had a relative risk of death of 0.42 ($p<0.001$). In a study combined analysis of two French Cooperative Thoracic Intergroup (the Intergroupe Francophone de Cancérologie Thoracique, IFCT) randomized trials,[26] 5-year OS was 80.0% in the PCR group, compared with 55.8% in the non-PCR ($p=0.0007$) and hazard ratios (HR) for death with PCR was 0.34 (95% CI, 0.18–0.64) by multivariate analysis.

However, the rarity of PCR in patients with cisplatin-based chemotherapy restricted was usually less than 10%. It was reported that[27] each percentage of viable tumor was associated with a 1% increase in the risk of death (HR, 1.01, $p=0.005$). In a follow-up study,[28] only pathological stage and viable tumor (≤10%) associated with OS (HR 2.39, $p=0.05$), therefore major pathological response (MPR), defined as 10% or less residual tumor tissue in resected lung and lymph node tissue, were proposed as a surrogate of OS in patients with resectable NSCLC given neoadjuvant chemotherapy.[29] As for neoadjuvant immunotherapies, MPR has been used as a primary or second endpoint in some researches.[30] Above experience about MPR comes from trials of neoadjuvant chemotherapies mostly, due to the different mechanisms of chemotherapy and immunotherapy, there are differences in pathological assessment between them. Histopathologic features of the

regression bed (the area of immune-mediated tumor clearance) were found in the pathological assessment of NSCLC patients with neoadjuvant nivolumab and were proposed to develop "Immune-Related Pathologic Response Criteria" (irPRC) that standardize pathologic assessment of immunotherapeutic efficacy, which add the area of the regression bed to the areas of residual viable tumor and necrosis and detailed terms "stroma," "fibrosis," and "inflammation" to include only proliferative fibrosis (vs old, hyalinized fibrosis or any fibrosis), dense (vs mild) tumor infiltrating lymphocytes, and tertiary lymphoid structures (vs non-organized lymphoid aggregates).[32] Long-term follow-up is needed to validate MPR assessed by as a surrogate for recurrence-free survival and OS in researches about neoadjuvant immunotherapies.

3. Clinical trials on neoadjuvant immune checkpoint inhibitors

Neoadjuvant therapies provide opportunities to implement preoperative smoking cessation and reduce tumor burden before surgery. It was reported that neoadjuvant immunotherapy with nivolumab was associated with little side effects and did not postpone the surgery.[30] Clinical trial LCMC3, with neoadjuvant atezolizumab (n=77), reported MPR rate was 19% and only 6 grade 3–4 treatment-related adverse reactions occurred in 101 patients. In a clinical trial with nivolumab (n=21),[30] MPR occurred amazingly in 9 of 20 completely resected NSCLC and the number of T-cell clones changed after PD-1 inhibitors in 8 of 9 patients. Neoadjuvant immune checkpoint inhibitors might play a key role in activating specific immune killing of tumor cancer before operation.

These excellent results from neoadjuvant mono immunotherapy stimulate interest in neoadjuvant immunotherapies combined with chemotherapy or other checkpoint inhibitors. In the NEOSTAR trial, MPR + PCR rate of group nivolumab plus ipilimumab is only 16% higher than group nivolumab, but combined therapy significantly reduced the chance of subsequent surgical treatment (2 in group nivolumab vs 5 in the combined group). Moreover, trial CheckMate-617 about neoadjuvant combined checkpoint inhibitors was completely terminated in an early stage. Even so, trial NADIM about neoadjuvant nivolumab + chemotherapies showed an excellent result that MPR rate reached 83% and PCR rate reached 71%, but the trial is a small sample (n=46) research, next, more and more trials would be needed for exploring the best dose, circle, and combination for neoadjuvant immune checkpoint inhibitors. Recently, many clinical trials about neoadjuvant immune checkpoint inhibitors are ongoing (Table 2). We can expect them to bring exciting results.

Predictive biomarkers for adjuvant and neoadjuvant immunotherapies

An efficient predictive biomarker will be specific for patients' election in the clinical trial of neo- and adjuvant immune checkpoint inhibitors. As the proposed detection item by the Food and Drug Administration (FDA), PD-L1 on tumor cell is suggested to be a biomarker for anti-PD-1 inhibitor. PD-L1 protein expression assessed by immunochemistry (IHC) has emerged as a biomarker to select NSCLC patients for pembrolizumab therapy.[14,33,34] Moreover, Zaric et al[35] reported that PD-1 expression was an independent prognostic factor for recurrence and death, which revealed that PD-1 and PD-L1 expression were associated with favorable OS in patients with completely resected adenocarcinoma of the lung. However, Tsao et al[36] held a different opinion and showed that PD-L1 protein expression was not a prognostic factor in early-stage NSCLC patients. PD-L1 expression as an effective predictor to select patients with lung cancer for neo- and adjuvant immunotherapies needs to be further explored.

High tumor mutation burden (TMB), an emerging biomarker for response to immunotherapy, means the total number of mutations present in a tumor specimen.[37] TMB was first associated with clinical benefit in melanoma patients treated with anti-cytotoxic T lymphocyte associated antigen-4 (CTLA-4).[38] Afterward, Owada-Ozaki et al[39] found TMB >62 was associated with shorter OS (HR=12.31, p=0.019) in patients with resected NSCLC, while Roszik et al[40] reported that high TMB group treated with ipilimumab were correlated with better prognosis (HR=0.272, p=0.003). Moreover, it was reported that the rate of MPR has no significant difference between PD-L1–positive and PD-L1–negative tumors, but a significantly higher mean mutational burden was observed in tumors with an MPR than in tumors without a major response.[30] Therefore, TMB is a potential predictive biomarker for MPR following adjuvant and neoadjuvant immunotherapies.

Liquid biopsy

Liquid biopsy is a promising tool for noninvasive monitoring response in neoadjuvant or adjuvant immunotherapies. Circulating tumor DNA (ctDNA) appears to be present in

Table 2 Clinical trials of neoadjuvant immune checkpoint inhibitors

Study name (or Identifier)	Phase of trial	Drugs	Sample size	Stages	MPR rate (n/N)	PCR rate (n/N)
CheckMate-159	2	Nivolumab	20	I-IIIA	45% (9/20)	10% (2/20)
LCMC3	2	Atezolizumab	101	IB-IIIB	18% (15/82)	5% (4/82)
NEOSTAR	2	Nivolumab or nivolumab + ipilimumab	44	I-IIIA	24% (10/41)	15% (6/41)
NADIM	2	Nivolumab + carboplatin + paclitaxel	46	IIIA	83% (34/41)	71% (29/41)
KEYNOTE-671	3	Pembrolizumab + chemotherapy vs chemotherapy only	786 (Estimated)	IIB, IIIA	Ongoing	Ongoing
IMpower030	3	Atezolizumab + chemotherapy	374 (Estimated)	II, IIIA, or select IIIB	Ongoing	Ongoing
NCT03732664	1	Nivolumab	40 (Estimated)	IA3-IIIA	Ongoing	Ongoing
CheckMate 816	3	Nivolumab + ipilimumab or chemotherapy vs chemotherapy only	350 (Estimated)	IB-IIIA	Ongoing	Ongoing
AEGEAN	3	Durvalumab + chemotherapy vs chemotherapy only	300 (Estimated)	II, III	Ongoing	Ongoing
NeoCOAST	2	Durvalumab or durvalumab + oleclumab or monalizumab or danvatirsen	160 (Estimated)	I [>2 cm] to IIIA	Ongoing	Ongoing
CANOPY-N	2	Canakinumab or pembrolizumab or combination	110 (Estimated)	IB-IIIA	Ongoing	Ongoing

Abbreviations: MPR, major pathological response; PCR, Pathological complete response.

50–95% of patients with stages I through III,[41–43] suggesting it may be a more broadly applicable biomarker in this setting. Moreover, immunotherapies could cause dramatic activation in blood CD4(+) and CD8(+) T cells in some researches.[44] Updated data from trials CA209-159 also suggested that ctDNA clearance and peripheral blood T cell amplification may be potential predictors of therapeutic response and monitoring recurrence. However, it's still a question whether a change of ctDNA and peripheral blood T cell correlate with MPR, even with OS or DFS. Moreover, blood collection procedures, collection tubes, anticoagulant,[45] blood storage condition, blood centrifugation speed for plasma isolation,[46] and plasma storage condition[47] are also limiting factors associated with the standardization of circulating tumor DNA (ctDNA) to the clinical practice. To explore the clinical utility of these assays in patients receiving adjuvant and neoadjuvant immunotherapy, future trials should include serial sample collection for liquid biopsies.

Challenges and prospects

Lung cancer is the most commonly diagnosed cancer with the highest rate of death. Even for early stage resectable NSCLC, the rate of recurring or death is more than 8%. Recently, immune checkpoint inhibitors are hotspots in the treatment of cancer, but the best timing of immunotherapies use still need to explored. Neoadjuvant and adjuvant immunotherapies attached researchers' attention for some excellent results. Due to the particularity of treatment, immunotherapies will have bright prospects as neoadjuvant and adjuvant therapies, but some challenges have to be faced.

First of all, compared to chemotherapies, neoadjuvant and adjuvant immunotherapies have significant survival beneficial, but the use of immunotherapies has a risk of causing autoimmune disease, especially for neoadjuvant therapies, which causes patients to be unable to undergo surgery. Moreover, it should be considered that premature use of immunotherapies in early-stage lung cancer would increase immunotherapies drug resistance occurs in advance. Especially for neoadjuvant immunotherapies, it needs more exploration whether neoadjuvant immunotherapy will aggravate the specific problems of clinical practice such as adhesion and hemorrhage during operation, increase the difficulty of surgery and prolong the time of thoracic drainage.

Secondly, exploration about pathological response for neoadjuvant immunotherapies is a very worthwhile

challenge. As a surrogate for recurrence-free survival and OS, it can help researchers greatly reduce research time. However, the prerequisite for MPR to be used in clinical practice is to be able to predict the patient's OS. Long-term follow-up is needed to validate MPR as a surrogate for recurrence-free survival and OS in NSCLC researches about neoadjuvant immunotherapies.

Thirdly, neoadjuvant and adjuvant immunotherapies are still in the start-up stage, the dose and circles are both in the exploration. In current clinical trials, mono immunotherapies, immunotherapies + chemotherapies and immunotherapies + immunotherapies are commonly used programs. The combination of the anti-CTLA-4 antibody ipilimumab with either nivolumab or pembrolizumab has shown to have higher response rates than anti-PD-1 monotherapy, but at the cost of significant toxicity.[48,49] More recently, immunotherapies + chemotherapies (the trials NEOSTAR) showed best results, however, more clinical trials with large sample size are needed to verify. Moreover, whether neoadjuvant combined adjuvant immunotherapies or mono neoadjuvant or adjuvant immunotherapies are better, it's also a challenge that we need to explore.

Last, researchers have long wanted to screen for appropriate patients through molecular markers. To date, there are some promising indicators, however, whether PD-L1, TMB or recently emerging liquid biopsy (ctDNA, peripheral blood T cell and so on), there is not sufficient evidence to prove that they are directly related to MPR or OS. It's still controversial to screen for appropriate patients through these markers. In future research, according to the characteristics of immunotherapy itself, the most important object is to develop a comprehensive index that can reflect both the oncological response and the immunological response. Only in this way can we truly predict the immunotherapies' effect in real time.

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Disclosure

The authors report no conflicts of interest in this work.

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