Review Article

Narrative review: blood and tumor biomarker testing in non-small cell lung cancer without an oncogenic driver

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Background and Objective: For patients with metastatic non-small cell lung cancer (NSCLC) without an oncogenic driver, systemic therapy with immune checkpoint inhibitors (ICIs) alone or in combination with chemotherapy have significantly improved the outcomes. However, the majority of patients do not have a durable response, and there is a need for additional predictive biomarkers. The objective of this narrative review is to describe potential biomarkers for immunotherapy.

Methods: Narrative overview of the literature synthesizing the findings of literature reporting retrospective, prospective, and subset analyses of studies investigating potential predictive biomarkers for ICI.

Key Content and Findings: Tumor expression of programmed death ligand-1 (PD-L1) is the only clinically available biomarker for patients receiving ICI-based therapy. However, PD-L1 has significant limitations and studies have investigated the predictive value of higher PD-L1 expression levels. There has been interest in tumor mutation burden (TMB) based on the premise that a higher TMB would be associated with a more neoantigens, which would increase the likelihood of an immune response. The studies to date have not revealed a consistent association with TMB level and survival benefit. Kelch-like ECH Associated Protein 1 (*KEAP1*) and serine/threonine kinase 11 (*STK11*) mutations have been associated with worse outcomes with ICI but these mutations appear to be associated with a worse prognosis, and not predictive for ICI. Tumor infiltrating lymphocytes (TIL's) are the mechanism of immune response, and there is interest in further investigating the presence, type and distribution of TIL's to predict immune benefit. Circulating tumor deoxyribonucleic acid (ctDNA) levels, at baseline and on treatment samples, are being investigated to assess response to therapy and long-term benefit of ICI.

Conclusions: None of the current biomarkers in development are validated for use in routine clinical care. Given the complexity of NSCLC biology and immune response to ICI most likely a composite biomarker using multiple biomarkers will need to be develop.

Keywords: Immune check point inhibitors; circulating tumor DNA (ctDNA); programmed death ligand-1 (PD-L1); tumor mutational burden (TMB)

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Introduction

Lung cancer is the leading cause of cancer mortality in the United Sates and a leading cause globally, and the majority of patients have the non-small cell lung cancer (NSCLC) subtype (1-3). The majority of patients with NSCLC have locally advanced or metastatic disease at the time of diagnosis, and the primary therapy is systemic therapy. Historically, the standard therapy for patients with metastatic NSCLC was platinum-based chemotherapy, which improved overall survival (OS) modestly. The thoracic oncology field recognized that platinum-based chemotherapy had reached a therapeutic plateau, and focused on the development of biologic and targeted

Table 1 Search strategy summary

Item	Specification
Date of search	March 2022 to June 2022
Data base	PubMed
Search terms	PD-L1, tumor mutation burden, *KEAP1* and *STK11* mutations, tumor infiltrating lymphocytes, and circulating tumor DNA, immunotherapy or immune checkpoint inhibitors
Timeframe	Between January 2000 to June 2022 with the full publication
Inclusion and exclusion criteria	Inclusion criteria: peer review clinical studies available in English
	Exclusion criteria: preclinical studies, secondary publications, review articles, editorials or commentaries, case reports, case series
Selection process	Priority given to primary publication rather than secondary publications of clinical trials or long-term follow-up studies
Additional considerations	Focus on biomarker studies with clinical outcomes

PD-L1, programmed death ligand-1.

therapies (4).

The initial studies of targeted therapies were conducted in patients without biomarker selection criteria; however, in the trials of epidermal growth factor receptor (EGFR) tyrosine kinase inhibitors (TKIs) patients with certain clinical characteristics were observed to have a higher response rate. These observations led to the identification of *EGFR* sensitizing mutations, and subsequent trials demonstrated the superiority of EGFR TKIs compared to platinum-based chemotherapy in patients with *EGFR* mutant NSCLC (5,6). This ushered in the era of precision medicine, the development of multiple targeted therapies in patients with a specific biomarker which improved the outcomes of this subset of patients with "oncogenic driven" NSCLC. This also led the division of NSCLC into the clinical categories of "oncogenic driven" and "non-oncogenic driven" NSCLC.

The next major therapeutic advance in the treatment of metastatic NSCLC was the development of immune checkpoint inhibitors (ICIs). The initial trials revealed activity in patients who had received multiple lines of therapy, and there was trend for greater benefit with higher programmed death ligand-1 (PD-L1) tumor expression. Trials of ICI monotherapy revealed less benefit for patients with *EGFR* mutant and anaplastic lymphoma kinase (*ALK*) rearranged NSCLC, which led to the exclusion of these NSCLC subtypes from ICI trials. Subsequent trials in the first-line setting established the superiority of ICI monotherapy compared to chemotherapy in patients selected based on PD-L1 tumor expression, and chemotherapy and ICI combinations compared to chemotherapy alone regardless of the level of PD-L1 expression (7). Clinically, patients are described as PD-L1 low (PD-L1 <1%), intermediate (PD-L1 1–49%), and high (PD-L1 ≥50%) to assist with selecting ICI monotherapy or chemotherapy and ICI combinations (8-12).

For patients without an oncogenic driver the standard therapy is ICI monotherapy or in combination with chemotherapy. However, many patients do not experience a response or durable benefit from ICI containing therapy. Thus, there is a clinical need to develop better predictive biomarkers of ICI benefit. The role of circulating tumor deoxyribonucleic acid (ctDNA) levels at baseline and with serial ctDNA levels to predict durable benefit from ICI is another an area of active investigation. This article is presented in accordance with the Narrative Review reporting checklist (available at https://tlcr.amegroups.com/article/view/10.21037/tlcr-22-530/rc).

Methods

A literature review using publication PubMed (https://pubmed.ncbi.nlm.nih.gov/) to identify recent studies and literature reviews of biomarkers for immunotherapy using the specific biomarker term (*Table 1*). Studies published in peer reviewed journals were used. Specific terms used include PD-L1, tumor mutation burden (TMB), Kelch-like ECH Associated Protein 1 (*KEAP1*) and serine/threonine kinase 11 (*STK11*) mutations, tumor infiltrating lymphocytes (TIL's), and circulating tumor DNA in

Table 2 Outcomes of patients with PD-L1 ≥50% receiving immune checkpoint inhibitors as monotherapy: retrospective study of first line pembrolizumab (19)

PD-L1 cohort	Sample size	ORR (%)	Median PFS (months)	Median OS (months)
50–89%	80	33	4.1	15.9
90–100%	107	60	14.5	NR
Comparison		P<0.001	HR: 0.50, P<0.01	HR: 0.39, P=0.002

PD-L1, programmed death ligand-1; ORR, objective response rate; PFS, progression-free survival; OS, overall survival; HR, hazard ratio; CI, confidence interval; NR, not reported.

association with immunotherapy or ICIs.

PD-L1 expression

PD-1 is an inhibitory receptor expressed on T-cells and mediates immunosuppression, and tumor cells can express the ligands PD-L1 and programmed death ligand 2 (PD-L2). The interaction of PD-1 with the PD-L1 ligand inhibits the immune response, and inhibition of this inhibitory interaction can induce T-cell response and immune response to tumor antigens (13). This is a complex pathway and the PD-1/PD-L1 interaction is only one of the multiple pathways involved in immune response. The initial study of nivolumab revealed an association between objective response rate (ORR) and expression of PD-L1 using immunohistochemistry (IHC) testing (14). Subsequently, several different IHC assays were developed in conjunction with a specific ICI agent, which created a debate about the whether the performance of the assays were comparable. When the assays were assessed several performed similarly (22C3, 28-8 and SP263 assays), and the SP142 assay was less sensitive (15). Additional issues identified were the temporal and spatial and intra-tumoral heterogeneity of PD-L1 expression, specimen quality, and biopsy location influencing PD-L1 expression levels (16,17).

The development of pembrolizumab was associated with tumor proportion score (TPS) cut-offs using anti-PD-L1 antibody clone 22C3, and the regulatory approval of pembrolizumab was defined by specific PD-L1 expression levels (18). The co-development of the 22C3 assay and pembrolizumab led to the common use of the 22C3 assay. In the initial study of pembrolizumab in NSCLC, a receiver operating characteristic analysis revealed a TPS score of ≥50% as the cutoff. The ORR among patients with TPS PD-L1 ≥50%, 1–49%, and <1% was 45.2%, 16.5%, and 10.7%, respectively (18). A phase 3 study revealed superior overall survival (OS) with pembrolizumab compared to chemotherapy in treatment naïve patients with a TPS of ≥50%, and a subsequent study revealed superior OS with pembrolizumab compared to chemotherapy in treatment naïve patients with TPS of ≥1% (8,12). These studies established the minimal TPS expression for treatment with pembrolizumab monotherapy. Subsequent trials demonstrated the activity of atezolizumab and cemiplimab in patients with PD-L1 expression of ≥50% (10,11).

While the use of PD-L1 expression to select patients has become standard in clinical care, there are some fundamental limitations as a biomarker. The ORR in the "high" expression group is below 50%, and there have been subsequent investigations to assess if a higher threshold would result in higher ORR (12). The first study was a retrospective study investigating the outcomes among patients who received first-line pembrolizumab with tumor PD-L1 ≥50%, and who were negative for genomic alterations in the *EGFR* and *ALK* alterations. Patients with tumor PD-L1 expression ≥90% compared to 50-89% had a statistically significant higher ORR, longer progression-free survival (PFS) and OS (*Table 2*) (19). A *post-hoc* analysis of the phase 3 trial of cemiplimab compared to chemotherapy revealed a similar association between higher PD-L1 expression and better outcomes as well (*Table 3*) (10). The most logical explanation for these results is the relationship between PD-L1 expression is linear. Despite these results, it is unlikely we will have prospective studies investigating for better outcomes with PD-L1 expression ≥50% because with the development of chemotherapy and ICI combinations we are less reliant of PD-L1 expression for selection of ICI.

Additional data about the performance of PD-L1 assays are available from clinical trials. The phase 3 trial of atezolizumab compared to chemotherapy assessed PD-L1 expression in a *post-hoc* analysis using the SP142, 22C3, and SP263 assays. Variation in the concordance high expression was observed among the assays. However, the hazard ratio (HR) for OS in the high expression on the 22C3, SP142,

Table 3 Post-hoc analysis of the phase 3 trial of cemiplimab compared to chemotherapy (10)

PD-L1 cohort	Sample size	ORR (%)	Median PFS (months)	Median OS (months)
<50% or unknown	147	26 *vs.* 22	4.1 *vs.* 5.0; HR: 0.82 (95% CI: 0.56–1.18)	16.5 *vs.* 15.2; HR: 1.082 (95% CI: 0.68–1.72)
≥50%–≤60%	192	32 *vs.* 23	4.3 *vs.* 6.2; HR: 0.79 (95% CI: 0.56–1.12)	21.9 *vs.* 14.0; HR: 0.77 (95% CI: 0.49–1.23)
>60%–<90%	179	39 *vs.* 20	6.2 *vs.* 4.2; HR: 0.55 (95% CI: 0.38–0.80)	22.1 *vs.* 12.0; HR: 0.47 (95% CI: 0.27–0.80)
≥90%	192	46 *vs.* 18	15.3 *vs.* 5.9; HR: 0.28 (95% CI: 0.17–0.46)	NR *vs.* 15.1; HR: 0.46 (95% CI: 0.25–0.85)

PD-L1, programmed death ligand; ORR, objective response rate; PFS, progression-free survival; OS, overall survival; HR, hazard ratio; CI, confidence interval; NR, not reported.

and SP263 assays were 0.60 [95% confidence interval (CI): 0.42 to 0.86], 0.59 (95% CI: 0.40 to 0.89), and 0.71 (95% CI: 0.50 to 1.00), respectively. In the phase 3 trial of cemiplimab, the tumor expression was assessed using the 22C3 assay at central laboratory, and study monitoring revealed that assay was not consistently analyzed per instructions. This inconsistency created a study patient population of 700 patients and a patient population of 563 patients with PD-L1 expression ≥50% per prescribed instructions by the test manufacturer. The data from the cemiplimab clinical trial raises the concern that variation in methods and assessment of PD-L1 expression may be an issue in routine clinical care. These analyses from phase 3 trials do not suggest that the predictive value of PD-L1 expression for OS will be improved by using a different assay.

TMB

Early studies demonstrated an association between high TMB and benefit from ICI in multiple tumor types, and the scientific rationale was that tumors with a higher TMB have a higher number of tumor neoantigens, thus have a higher probability of inducing an immune response (20,21). The measurement of TMB is defined as the number of nonsynonymous mutations per coding area of the genome (22). TMB and PD-L1 expression were observed to be independent biomarkers (23). There was hope the TMB level could be used as an additional predictive marker of ICI benefit in NSCLC. Different testing methods have been used to determine the TMB, and this has required harmonization of the testing methods (24).

In the phase 3 trial of nivolumab compared to chemotherapy, the association between clinical benefit and TMB level was assessed in an exploratory analysis, and 50% of the enrolled patients were included in the analysis. Patients in the TMB high category experienced a higher ORR and longer PFS, and there was not an association between TMB and PD-L1 expression. Data from a single arm phase 2 trial of nivolumab and ipilimumab demonstrated TMB as a potential biomarker and established the cut-off of TMB 10 mutations/megabase (Mb). The subsequent phase 3 trial of nivolumab and ipilimumab compared to chemotherapy prospectively evaluated PFS in the subset of patients with TMB ≥10 mutations/Mb (25,26). Of the 1,739 randomly assigned patients, 1,649 (95%) had tumor samples available for TMB analysis, and 1,004 (58%) had valid data for TMB and were included in the efficacy analysis. Patients with a higher TMB had a higher ORR and longer PFS (*Table 4*); however, the OS benefit was similar in the TMB <10 mutations/Mb subset (HR 0.75, 95% CI: 0.59–0.94), and TMB ≥10 mutations/Mb subsets (HR 0.68, 95% CI: 0.51–0.91). These results raised concerns about the predictive value of TMB for OS benefit. However, a number of factors could have contributed to the results including trial design issues since this was a subset analysis, a potential interaction between a prognostic and predictive value of TMB, and the selection of the cut-off value (29). Furthermore, the association between tumor mutation and neoantigen is complex and some mutations may produce more or less potent tumor neoantigens (30).

One inherent limitation to tumor TMB testing is that some patients have insufficient tumor tissue for TMB testing, and the turnaround time can be problematic when patients need to initiate therapy. There had been increasing interest in using blood TMB (bTMB) to select patients for immunotherapy. A blood-based assay was able to define patients bTMB and develop cut-offs using retrospective data from randomized trials of single agent ICI (31). In a phase 3 trial of durvalumab, durvalumab and tremelimumab, and chemotherapy there was an exploratory analyses bTMB level and outcome, and 72% of the enrolled patients were evaluable for bTMB testing (27). A numerically higher ORR, longer PFS and OS was observed in the subset of patients with bTMB ≥20 who received durvalumab and

Table 4 Select first-line trials evaluating efficacy of immune checkpoint inhibitors based on TMB level

First author	Comparison	Subset	ORR	PFS	OS
Hellman (26)	Nivolumab/ipilimumab *vs.* chemotherapy	TMB ≥10 (n=299)	45.3%; 26.9%	HR: 0.58; 97.5% CI: 0.41–0.81	HR: 0.68; 95% CI: 0.51–0.91
		TMB <10 (n=380)	NR	HR: 1.07; 95% CI: 0.84–1.35	HR: 0.75; 95% CI: 0.59–0.94
Rizvi (27)	Durvalumab/tremelimumab *vs.* chemotherapy	bTMB ≥20 (n=134)	48.4%; 21.4%	HR: 0.53; 95% CI: 0.34–0.81	HR: 0.49; 95% CI: 0.32–0.74
		bTMB <20 (n=389)	16.7%; 31.4%	HR: 1.55; 95% CI: 1.23–1.94	HR: 1.16; 95% CI: 0.93–1.45
	Durvalumab *vs.* chemotherapy	bTMB ≥20 (n=147)	29.9%; 21.4%	HR: 0.77; 95% CI: 0.52–1.13	HR: 0.72; 95% CI: 0.50–1.05
		bTMB <20 (n=389)	20.6%; 31.4%	HR: 1.19; 95% CI: 0.94–1.50	HR: 0.93; 95% CI: 0.74–1.16
Peters (28)	Atezolizumab *vs.* chemotherapy	bTMB ≥16 (n=472)	25.5%; 17.8%	HR: 0.77; 95% CI: 0.59–1.00	HR: 0.87; 95% CI: 0.64–1.17

TMB, tumor mutation burden; bTMB, blood TMB; ORR, objective response rate; PFS, progression-free survival; OS, overall survival; HR, hazard ratio; CI, confidence interval; NR, not reported.

tremelimumab compared to chemotherapy (*Table 4*). The combination of durvalumab and tremelimumab was compared to platinum-based chemotherapy in patients with high bTMB (≥20) in a prospective study and the trial did not demonstrate an improvement in OS (28,32,33). The Blood First Assay Screening Trial (BFAST) cohort prospectively assessed atezolizumab in patients with high bTMB. The primary end-point was PFS in the patient cohort with bTMB ≥16, and the trial did not demonstrate an improvement in PFS (*Table 4*). In an exploratory analysis, the HR for PFS improved numerically with a higher bTMB threshold but the OS outcomes did not improve with higher bTMB thresholds.

One fundamental question in the studies evaluating TMB is the optimal cut-off. A recent retrospective study examined the optimal cut-off using multiple patients cohorts, and determined that a TMB level >19 was associated with better outcomes from ICI (34). Of the 1,552 patients in the cohort, 161 patients (10%) had TMB >19.0 mutations/Mb, and 1,391 patients (90%) had a TMB ≤19 mutations/Mb. For patients with a TMB >19.0 compared to patients with TMB ≤19 treated with ICI, a higher ORR (42.5% *vs.* 18.0%, P<0.001), longer PFS (HR of 0.38, 95% CI: 0.28–0.52, P<0.001) and longer OS (HR of 0.46, 95% CI: 0.32–0.65, P<0.001) were observed. These results were consistent among patients with a PD-L1 expression of <1%, 1–49%, and ≥50%.

The aggregate data demonstrate a trend towards better outcomes with ICI for patients with higher TMB levels. Of note, many patients in the TMB low category have oncogenic driven NSCLC, which has been associated with less benefit from ICI containing therapy, and this patient population may be contributing to the results in the TMB low category. However, the studies to date are not definitive and TMB is an intriguing biomarker but not validated for routine clinical use in NSCLC. The future role for TMB testing will most likely be to identify patients with low PD-L1 and high TMB who could benefit from ICI monotherapy

KEAP1 and STK11 mutations

Recently, there has been interest in assessing if specific mutations may be associated with ICI response or resistance based on preclinical and retrospective clinical data. The recent focus has been on *KEAP1* and *STK11* mutations, and the prevalence of *KEAP1* and *STK11* mutations is approximately 20% (35). The KEAP1-NRF2 system is a defensive mechanism against oxidative and electrophilic stresses, and *KEAP1* mutations are associated with resistance to chemotherapy and radiation therapy (36). Retrospective data have suggested that patients with tumors with *KEAP1* single mutation have a worse prognosis with ICI compared to patients with tumors with *KEAP1/TP53* co-mutations or *TP53* mutation alone (37). Additional studies have revealed the presence of other co-mutations [*STK11*, *SMARCA4* or protein polybromo-1 (*PBRM1*)] are associated with worse outcomes compared to single mutation or wild-type (36,38). *KEAP1* mutations are associated with worse outcomes

among patients with Kirsten rat sarcoma virus (*KRAS*) mutant NSCLC, but not patients with *KRAS* wild-type NSCLC (35).

STK11 (also known as *LKB1*) regulates cellular metabolism and growth, *STK11* mutant NSCLC is associated with lower PD-L1 expression, lower T-cell infiltration, and less benefit from ICI. The mechanisms is due to silencing of the stimulator of interferon genes (STING) expression (39,40). Similar to *KEAP1* mutations, *STK11* mutations confer worse outcomes among patients with concurrent *KRAS* mutations and not *KRAS* wild-type (35).

A retrospective, real world study investigated the outcomes of 2,276 patients including 574 patients treated with ICI as first-line therapy (41). *STK11* and *KEAP1* mutations alone or in combination were associated with worse outcomes with chemotherapy or ICI. An interaction between *STK11* and *KEAP1* and ICI on real world PFS and OS was not observed. Unfortunately, data from subset analyses from phase 3 trials are limited by small sample size, the retrospective design and the potential imbalance of other prognostic factors (42).

In summary, the presence of *KEAP1* or *STK11* mutation has been associated with a worse prognosis, and there is a preclinical rationale for this subgroup of patients to be less responsive to ICI. However, the data to date suggest that these mutations may be prognostic rather than predictive biomarkers. The confounding factor of concurrent mutations makes assessment of the individual mutations difficult. A prospective study to evaluate the predictive role of these mutations and concurrent mutations or prospective collection of mutation status, therapy, and outcomes in a registry would be required to assess the role these mutations.

TIL's

TIL's are the primary mechanism of immune response to malignancies, and there has been interest in defining TIL's as a prognostic and predictive marker for NSCLC. Previous studies had demonstrated that the presence of TIL's was associated with a better prognosis, including in early stage NSCLC (43,44). In addition to the presence or absence of TILs, the specific TIL phenotype, and spatial distribution may contribute to the outcome. With the development of ICI there was renewed interest in assessing TIL's as potential complementary biomarker of ICI benefit. Tumors are frequently defined as inflamed (TIL distribution intra-tumorally), immune excluded (TIL excluded, outside of the cancer stroma), and immune desert (scant TIL in the tumor microenvironment) (45).

Some of the challenges in developing TIL's as biomarker is that identification and quantitation of TIL's is labor intensive and inter-observer heterogeneity can be an issue (46). A study using artificial intelligence-powered spatial analysis of TIL's, and assessed the outcomes of patients with immune inflamed and non-inflamed tumors with ICI. Of the tumors, 44%, 37%, and 19% were immune inflamed, immune excluded, and immune desert, respectively, and the rate of inflamed tumors increased with increasing PD-L1 levels. Patients received single agent ICI, and the ORR in the inflamed, immune excluded, and immune desert was 26.8%, 11.5%, and 11.2%. The median PFS in the immune inflamed, immune excluded, and immune desert was 4.1, 2.2 and 2.4, respectively (HR 1.52, P<0.001 for immune-excluded *vs.* inflamed and 1.58, P<0.001 for immune-desert *vs.* inflamed). The median OS in the immune inflamed, immune excluded, and immune desert was 24.8, 14.0 and 10.6 months, respectively (HR 1.38, P<0.05 for immune-excluded *vs.* inflamed and 1.67, P<0.05 for immune-desert *vs.* inflamed). A recent study investigated the TIL's patterns in NSCLC adenocarcinoma and NSCLC squamous (47). A machine-learning model was used to assess the morphologic and molecular differences in immune patterns on digitized images of hematoxylin and eosin stains. The TIL signature differed between the histological subtypes, and the signature was prognostic in both subtypes. Samples from the phase 3 trial of nivolumab compared to docetaxel in patients with non-squamous NSCLC were analyzed, and in a retrospective analysis TIL density was associated with response to nivolumab but not docetaxel (48).

While the results from both of these studies are preliminary, they illustrate how computer-based analyses can be used to assess TIL's patterns and phenotypes in NSCLC, which historically has been a time and labor consuming process. Future studies are needed to better assess the clinical utility of TIL's as complementary predictive marker.

ctDNA

The use of ctDNA has revolutionized the management of metastatic NSCLC, and the primary use is to evaluate for oncogenic driver alterations which have an available targeted therapy (49). Several studies have investigated ctDNA as prognostic marker, either baseline or reduction in ctDNA levels. Patients with higher metastatic burden

or extra-thoracic metastases are associated with a greater chance of having detectable ctDNA, and a worse prognosis. Several analyses have investigated the outcomes of patients without oncogenic driver alterations receiving immunotherapy. A study involving multiple cancer types investigated the role of ctDNA from trials of durvalumab alone or with tremelimumab (50). Pre-treatment and on-treatment samples were available in 978 and 171 patients, respectively, and ctDNA was detectable on 83% of patients. In the NSCLC cohort (n=333) pretreatment levels below the median were associated with improved survival (HR of 0.66, 95% CI: 0.49–0.88). Pre-treatment levels were not associated with response. Paired ctDNA samples were available from subset of 171 patients (17.5%), and on-treatment ctDNA levels were associated with ORR. Patients were subdivided into 3 cohorts: increased ctDNA (n=75), decreased but not completely resolved ctDNA (n=68), and complete clearance of ctDNA (n=28). Statistically significant differences in PFS and OS were observed between these cohorts. An additional study of 62 patients receiving first line therapy with pembrolizumab alone or in combination with carboplatin and pemetrexed assessed outcomes of patients based on ctDNA levels 21 days after starting therapy (51). A reduction in ctDNA levels was associated with a higher response rate (60.7% *vs.* 5.8%, P=0.0003), longer PFS (HR of 0.29, 95% CI: 0.14–0.60, P=0.0007; median 8.3 and 3.4 months, respectively) and OS (HR of 0.34, 95% CI: 0.15–0.75, P=0.008; median 26.2 and 13.2 months, respectively). A smaller study of 28 patients revealed strong agreement between reduction in ctDNA and radiographic response, and longer PFS (HR of 0.29, 95% CI: 0.09–0.89, P=0.03) and OS (HR of 0.17, 95% CI: 0.05–0.62, P=0.007). Other studies have revealed a similar relationship (52,53).

The association between baseline ctDNA and prognosis and reduction in ctDNA and benefit to immunotherapy has established by these studies. Clinically, the question is whether the reduction in ctDNA assessment provides incremental value over the currently available radiographic assessment. One limitation of the studies to date is that they have used different assays, time points for collection, and definition of reduction in ctDNA. The development of a standard definition of molecular response and the optimal time point for assessment would help in the design of future studies. One potential clinical utility may be in assessing patients who have an ambiguous radiographic response. Another potential clinical question is using the on treatment ctDNA assessment to identify a subset of patients who have undetectable DNA, and this group of patients may be candidates for trials investigating a shorter compared to a longer duration of ICI.

Conclusions

While there is clearly a high unmet need for better predictive biomarkers of benefit or lack of benefit to ICI-containing therapy a biomarker beyond PD-L1 expression is not available. Each of the biomarkers investigated has revealed potential but are not well enough validated to be incorporated into clinical care. The issues of confounding clinical and molecular factors make assessment of new biomarkers challenging. Real-world data provide hypothesis generating or preliminary evidence but is unlikely to have the necessary data collection and rigor to lead to validation of a new predictive biomarker. Registry data, in which a limit number of critical factors are prospectively collected, may be the best method of developing biomarkers, since the subset analyses from phase 3 trials are limited by small sample size. Most likely, a composite biomarker using multiple factors will be needed to be developed given the complexity of the underlying biology NSCLC and immune response from ICI.

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Footnote

Reporting Checklist: The author has completed the Narrative Review reporting checklist. Available at https://tlcr.amegroups.com/article/view/10.21037/tlcr-22-530/rc

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