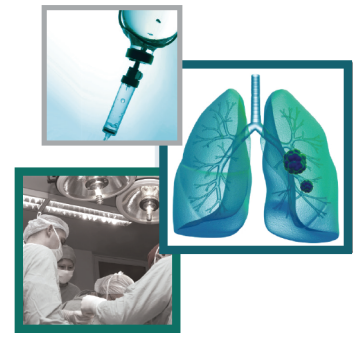
Editorial

Lung Cancer Management



Identifying successful biomarkers for patients with non-small-cell lung cancer

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"There are several new biomarkers in their infancy: the role and importance of TILs, immune gene signatures, interferon gamma related mRNA-based signatures, T-cell exhaustion profiling, myeloid-derived suppressor cells or the neutrophil-to-lymphocyte ratio at baseline, HLA diversity and the microbiota."

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Representing 11.6% of all cancers, lung cancer remains the most commonly diagnosed malignancy and the leading cause of cancer-related mortality worldwide, accounting for 18.4% of all cancer deaths [1]. It comprises two main subtypes, small cell and non-small-cell lung cancer (NSCLC), the latter representing over 80% of lung cancers. NSCLC can be further divided into two main histologic subtypes, adenocarcinoma (60%) and squamous cell carcinoma (35%) [2], each with separate mutational and genomic profiles.

Over the course of the past decade, a significant therapeutic breakthrough in the treatment of NSCLC has been the development of immune-checkpoint inhibitors (ICPI). The recent presentation of the 5-year overall survival (OS) of the Keynote 001 [3] at the ASCO 2019 meeting has further strengthened the importance and impact of ICPI on patients with NSCLC, showing durable long-term responses, and an unprecedented 5-year OS of 13.4–29.6% [4,5]. Although this is a radical improvement over the (<5%) 5-year survival previously seen in this setting, it still implies that roughly three-quarters of patients do not derive this degree of durable benefit from ICPI, despite still being exposed to the potential toxicities [6]. This leads to one of the major current challenges in oncology: the search for predictive biomarkers.

Developing biomarkers requires several steps: the first is to identify genes or proteins that are differentially expressed in tissues or fluids of specific groups of NSCLC patients. The sensitivity and specificity of these markers are then assessed. A successful biomarker is one that can predict the response, or lack thereof, of a tumor to a specific treatment.

Today, when it comes to ICPI in NSCLC, one biomarker stands out as having a proven clinical benefit; programmed-death ligand-1 (PD-L1). Assessed on tumor cells, the tumor microenvironment or a combination thereof, it allows for the identification of patients that are more likely to respond to PD-1 blockade. In the first-line management of NSCLC, patients with tumors expressing ≥50% PD-L1 have improved outcomes with pembrolizumab, a PD-1 ICPI, than with platinum doublet chemotherapy, both in terms of response rate (44.8 vs 27.8%) and OS [7]. Although this is the most clinically relevant biomarker today based upon available approvals, it has substantial limitations. It is disappointing that even among patients with substantial overexpression of PD-L1, over 50% of patients do not respond to pembrolizumab monotherapy.

Although PD-L1 staining is a relatively simple immunohistochemistry (IHC) assay, it is important to remember that each PD-1/PD-L1 inhibitor was developed alongside a separate PD-L1 staining assay. Although the IASLC Blueprint project showed that many of these assays are concordant on the tumors, there are outlier assays and the concordance on infiltrating immune stroma is much more limited [8]. However, even if we had completely concordant PD-L1 assays, it is important to remember that PD-L1 nonexpressers could still be responders and

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benefit from ICPI, meaning that it fails to rule out patients who should not receive these treatments. Conversely, in patients with NSCLC who are harboring driver mutations, the PD-L1 level can be misleadingly high, generally mediated by the JAK3 pathway. Despite this constitutional expression, these patients generally do not respond to ICPI [9], partially due to the tumor immune-microenvironment with a paucity of tumor infiltrating T-lymphocytes (TILs). Next, on a pre-analytic level, if PD-L1 is heavily glycosylated, it can lead to the absence of IHC staining. The performance of IHC assays can be improved through pre-analytic sample deglycosylation, which improves binding affinity, thus reducing false negatives [10]. Perhaps the most critical limitation of PD-L1 is that it is both dynamic and heterogeneous [11].

Given the clear limitations of PD-L1 staining, the need to identify and validate effective new biomarkers remains crucial. Many potential biomarker candidates are under investigation, with the goal of better tailoring treatments to patients and avoiding unnecessary toxicity.

A new promising biomarker is the tumor mutation burden (TMB). The prevalence of somatic mutation varies between 0.01 and 400 mutations/Mbp. Some of these mutations lead to the translation of novel peptide epitopes or neoantigens that could enhance the immunogenicity of the tumor by eliciting T-cell repertoire. The hypothesis is that, in cases of high TMB, ICPI should be more effective than chemotherapy. This hypothesis is supported by studies that have shown an improvement in response rate and progression-free survival, though no study has at this time confirmed an OS advantage in high-TMB patients [12]. However, in spite of promising early data and greater response rates, there appears to be no correlation between OS with single-agent ICPI and TMB in NSCLC, whereas TMB may have a predictive value when combining PD-1 blockade and anti-CTLA4 inhibition [13,14].

TMB also has some inherent technical issues that could dampen its clinical utility; the turnaround time for TMB is long, at least 2 weeks, and there is no assay harmonization, as TMB was historically evaluated on whole exome sequencing but has now shifted to next-generation analysis (NGS). Essentially, we do not know if NGS panel A concordant with whole exome sequencing would be concordant with NGS panel B. In addition, it entails a high cost, lacks uniform cut-offs with clinical implications and, given contradictory results, it is unclear whether TMB should be performed on cancer tissue or circulating tumor DNA. Again, all of these factors only become relevant if a prospective trial validates TMB as predictive of better OS for ICPI compared with standard of care, or in high TMB compared with low TMB.

There are several new biomarkers in their infancy: the role and importance of TILs, immune gene signatures, interferon gamma related mRNA-based signatures, T-cell exhaustion profiling, myeloid-derived suppressor cells or the neutrophil-to-lymphocyte ratio at baseline, HLA diversity and the microbiota. None of them are ready for prime time but there are ongoing studies with high hopes.

The question remains whether or not biomarkers for ICPI in NSCLC can become a reality. Given the impact of PD-L1, we believe they will. However, it is far less clear whether any of the established biomarkers will definitively address the known limitations. Although PD-L1 and TMB are orthogonal biomarkers, and those patients who are high for both have excellent outcomes, this represents a small subset of patients. All other groups of patients (high/low, low/high and low/low) are not substantially benefitted by combining PD-L1 and TMB. There is indeed room for improvement and more biomarkers are needed. Perhaps the answer does not lie in a single biomarker, but rather in a combination thereof, whether it could be put together as a panel with the support of artificial intelligence [15], an algorithm combining different clinical factors, as is the case in renal cancer, or a gene expression profile, as can be used in adjuvant breast cancer decisions. It could even be a combination of multiple complex parameters, aptly named multiomics. The latter is in its infancy but shows promise [16].

As important as a positive predictive biomarker is a negative one. There has been much enthusiasm pertaining to the novel identification of STK11 and KEAP1 tumor suppressor gene alterations as negative predictive biomarkers, suggesting the presence of these mutations led to an absence of response to ICPI in KRAS mutant NSCLC [17]. However, the data were retrospective and real-world analyses presented at the IASLC World Conference on Lung Cancer in September 2019 (Barcelona, Spain) did not support these hypotheses [18]. STK11 was correlated with poor response to first-line chemotherapy, but not second line, and to second-line immunotherapy. This suggests that these alterations may be prognostic rather than predictive. A prospective trial is necessary to confirm the impact of these mutations and evaluate their role as biomarkers in NSCLC.

In conclusion, the ideal characteristics required for a biomarker to be widely adopted in clinical practice are a short turnaround time, easily technical feasibility, reproducibility and, most importantly, the association with a proven OS association in prospective data. All of these would allow a biomarker for NSCLC to move from myth to reality.

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