Lung Cancer 144 (2020) 76–84

Contents lists available at ScienceDirect

Lung Cancer

journal homepage: www.elsevier.com/locate/lungcan

Review

Anti-angiogenic agents in the age of resistance to immune checkpoint inhibitors: Do they have a role in non-oncogene-addicted non-small cell lung cancer?

Sanjay Popat[a,b,*], Christian Grohé[c], Jesus Corral[d], Martin Reck[e], Silvia Novello[f], Maya Gottfried[g], Dejan Radonjic[h], Rolf Kaiser[h,i]

[a] Royal Marsden Hospital NHS Foundation Trust, 203 Fulham Road, Chelsea, London, SW3 6JJ, UK
[b] The Institute of Cancer Research, 15 Cotswold Road, Sutton, London, SM2 5NG, UK
[c] Department of Respiratory Diseases, ELK, 13125, Berlin, Germany
[d] Clínica Universidad de Navarra en Madrid, Calle Marquesado de Sta. Marta, 1, 28027 Madrid, Spain
[e] Department of Thoracic Oncology, Airway Research Center North (ARCN) Member of the German Center for Lung Research (DZL), LungenClinic, Wöhrendamm 80, 22927 Großhansdorf, Germany
[f] Department of Oncology, University of Turin, San Luigi Hospital, Regione Gonzole, 10, 10043 Orbassano TO, Turin, Italy
[g] Meir Medical Center, Tchernichovsky St 59, Kefar Sava, 4428164, Israel
[h] Boehringer Ingelheim International GmbH, Binger Strasse 173, 55216 Ingelheim am Rhein, Germany
[i] Institute of Pharmacology, Johannes Gutenberg-University Mainz, 55122 Mainz, Germany

ARTICLE INFO

Keywords:
Anti-angiogenic drug
Immunotherapy resistance
Nintedanib
Non-oncogene-addicted non-small cell lung cancer (NSCLC)
Tumor microenvironment (TME)
Vascular endothelial growth factor (VEGF)

ABSTRACT

The introduction of licensed front-line immunotherapies has heralded a new era for the treatment of non-oncogene-addicted, advanced non-small cell lung cancer (NSCLC). Yet as with all evolutions in clinical management, changes in practice can outpace the availability of the clinical evidence needed to inform subsequent therapeutic decision making. At the time of writing, there is limited available evidence on the optimum therapeutic options after progression on immunotherapy. Further research is needed to define mechanisms of immunotherapy resistance in patients with advanced NSCLC, and to understand the implications for subsequent treatment response. Pending the availability of robust clinical data and proven therapeutic options to underpin an optimized therapeutic pathway after progression on immunotherapy, attention must turn to the potential utility of currently licensed agents and any available supporting clinical data in this setting. Within this context we review the mechanistic arguments and supporting evidence for the use of anti-angiogenic agents as a means of targeting immunosuppression within the tumor microenvironment. We consider whether VEGF inhibition may help to normalize the tumor vasculature and to address immunosuppression – reinstating, and potentially enhancing, the effect of subsequent therapies. We also highlight evidence needs and signpost ongoing trials that should enable current clinical opinion in this area to be replaced by robust, evidence-based guidance.

Abbreviations: Ab, antibody; ABC, atezolizumab, bevacizumab and chemotherapy combination; ANG2, angiopoietin 2; APC, antigen-presenting cells; αPD-1, anti-programmed cell death protein 1; αPD-L1, anti-programmed death ligand-1; Arg1, arginase 1; β2M, beta-2 microglobulin; CCL, C-C-motif chemokine ligand; CI, confidence interval; CSF1, macrophage colony-stimulating factor 1; CTL, cytotoxic T lymphocyte; CTLA-4, cytotoxic T-lymphocyte antigen 4; CXCL12, C-X-C-motif chemokine ligand 12; DC, dendritic cell; DCR, disease control rate; EC, endothelial cell; ESMO, European Society of Medical Oncology; FASL, Fas ligand; GM-CSF, granulocyte–macrophage colony-stimulating factor; HLA, human leukocyte antigen; IDO, indolaimine-2, 3-deoxygenase; IFN, interferon; IL, interleukin; JAK, Janus kinase; LAG3, lymphocyte activation gene 3; M2 Mϕ, type II macrophage; MDSC, myeloid-derived suppressor cell; MHC, major histocompatibility complex; NF-κB, nuclear factor kappa B; NSCLC, non-small cell lung cancer; ORR, objective response rate; OS, overall survival; PD-1, programmed cell death protein 1; PD-L1, programmed death ligand-1; PD-L2, programmed death ligand-2; PFS, progression-free survival; PGE_2, prostaglandin E_2; PS, performance status; RECIST, Response Evaluation Criteria in Solid Tumors; STAT, signal transducer and activator of transcription; TAM, tumor-associated macrophage; TCR, T-cell receptor; T_{EM}, effector memory T cell; TGFβ, tumor growth factor beta; TGR, tumor growth rate; TIM3, T-cell immunoglobulin and mucin domain-3; TME, tumor microenvironment; TNF, tumor necrosis factor; T_{reg}, regulatory T cell; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor; VISTA, V-domain immunoglobulin suppressor of T-cell activation

* Corresponding author at: Royal Marsden Hospital NHS Foundation Trust, 203 Fulham Road, Chelsea, London, SW3 6JJ, UK.
E-mail address: Sanjay.Popat@rmh.nhs.uk (S. Popat).

https://doi.org/10.1016/j.lungcan.2020.04.009
Received 12 January 2020; Received in revised form 1 April 2020; Accepted 9 April 2020

1. Introduction

Despite the advent of public health initiatives in recent years, North America and Central and Eastern Europe continue to record some of the highest lung cancer incidence rates globally [1]. Non-small cell lung cancer (NSCLC) accounts for an estimated 80–90 % of all lung cancers, with adenocarcinomas representing an increasing healthcare priority across the USA and Europe [2].

The therapeutic landscape for stage IV NSCLC has been redefined in recent years with the advent of targeted agents directed at specific driver mutations and by the emergence of immunotherapies [2,3]. To reflect the rapidly expanding therapeutic armamentarium, national and international organizations, including the European Society of Medical Oncology (ESMO) and the National Cancer Comprehensive Network, have updated their recommendations to include immunotherapy strategies alongside traditional chemotherapy as front-line options (either as monotherapies in biomarker-selected patients, or in combination with chemotherapy) in eligible patients with advanced NSCLC without oncogenic driver mutations [2,3].

However, the speed at which immunotherapies have re-shaped the treatment landscape in this setting has resulted in a dearth of mature clinical data to guide treatment decisions for patients who progress on chemo-immunotherapy [2]. To facilitate such treatment decisions, we review a range of scenarios observed in this situation, outline mechanistic and clinical considerations that may influence subsequent treatment selection, and consider the potential role of anti-angiogenic agents within this rapidly changing therapeutic landscape.

2. Emerging clinical algorithms

In recent years, the development of agents targeting a wide range of oncogenic alterations (including *EGFR* and *BRAF* driver mutations, or gene rearrangements in *ALK* and *ROS-1*) has resulted in a clear strategy for treatment stratification according to the molecular profile of the tumor [2,4,5]. The emergence of immunotherapy has subsequently revolutionized therapeutic approaches for patients with NSCLC lacking such driver mutations [2], and has also shown potential in patients harboring oncogenic driver mutations once molecularly targeted options have been exhausted.

Checkpoint inhibitors targeting programmed cell death protein 1 (PD-1) and programmed death ligand-1 (PD-L1) have demonstrated durable disease response and prolonged survival in metastatic NSCLC. For example, patients with advanced, non-squamous NSCLC without sensitizing *EGFR* or *ALK* mutations treated with first-line pembrolizumab and chemotherapy achieved a response rate of 47.6 %, a median duration of response of 11.2 months, and a median overall survival (OS) of 22.0 months (hazard ratio 0.56; $p < 0.00001$ versus chemotherapy alone) [6,7]. Other PD-1- and PD-L1-targeted agents are now recommended by guideline bodies as first-line standard-of-care options alongside chemotherapy [2,3].

2.1. Patterns of clinical progression on immunotherapy

Despite the improved outcomes offered by immunotherapy as compared with chemotherapy alone, approximately half of patients with advanced NSCLC do not respond, and ultimately, almost all patients relapse. Although response is not the only marker for long-term treatment benefit with immunotherapy [8], disease progression often drives re-evaluation of the treatment plan. At the point of progression, many patients are eligible for additional systemic therapies, creating a growing need for evidence to guide sequential treatment options.

For patients with non-oncogene addicted NSCLC and PD-L1 expression ≥50 %, the 2019 ESMO guidelines recommend pembrolizumab monotherapy [2]; although it should be noted that there is some uncertainty regarding the decision to add chemotherapy based on the results from KEYNOTE-042 (pembrolizumab monotherapy: hazard ratio for OS, 0.69), KEYNOTE-189 (combination pembrolizumab plus chemotherapy: hazard ratio for OS, 0.59), and KEYNOTE-407 (pembrolizumab plus chemotherapy, squamous NSCLC: hazard ratio for OS, 0.64) [7,9,10]. Meanwhile, a growing body of evidence supports the addition of immunotherapy to chemotherapy for patients with newly diagnosed, metastatic NSCLC regardless of PD-L1 status (as shown by the OS results in KEYNOTE-189 and -407, in patients with PD-L1 expression 1–49 % or PD-L1 < 1 %) [2,7,10].

In patients who progress after chemotherapy ± immunotherapy, treatment with an anti-angiogenic plus docetaxel may be an option: either nintedanib (a small molecule drug targeting the tyrosine kinases vascular endothelial growth factor receptor [VEGFR] 1–3, platelet-derived growth factor receptor α and β, and fibroblast growth factor receptor 1–3) in patients with adenocarcinoma NSCLC, or ramucirumab (an anti-VEGFR2 antibody) in patients with performance status (PS) 0–2 [2].

However, the clinical utility of these second-line options has largely been inferred from trials conducted in the pre-immunotherapy era. For example, in the LUME-Lung 1 trial, patients with adenocarcinoma NSCLC who received nintedanib plus docetaxel after failure on first-line chemotherapy achieved a median OS of 12.6 months [11]. The REVEL trial of post-chemotherapy ramucirumab plus docetaxel in patients with NSCLC demonstrated a median OS of 10.5 months [12]. In the second-line, post-chemo-immunotherapy setting, there is a lack of high-quality evidence from prospective clinical trials to optimize second-line treatment selection. Further data on the impact of front-line chemo-immunotherapy and its implications for treatment sequencing are awaited. In the meantime, clinicians must take a pragmatic approach to sequential therapy selection, guided by determinants of potential treatment response as discussed below.

As the interactions between the immune system and cancer cells are continuous and evolve throughout periods of treatment [13], an important consideration when profiling patients progressing on combination chemo-immunotherapy is the nature of their initial response, and their pattern of progression (e.g. hyperprogression, pseudoprogression, or oligoprogression).

Drawing on the results of published immunotherapy trials, it is possible to categorize patients according to their response as: (i) 'responders' – individuals who initially respond and continue to experience clinical benefit; (ii) individuals with 'primary innate resistance' who appear to have no response and no benefit from treatment; and (iii) individuals with 'acquired resistance', who initially respond and derive benefit, but later acquire resistance and go on to relapse and progress [14].

Yet this apparently straightforward classification system obscures the complexity of the clinical reality. Response can differ spatially (between lesions) and also temporally – presenting immediately, rapidly, or as more slowly acquired resistance [14–17]. Moreover, the dynamic and constantly evolving nature of the immune response at the individual patient level can be influenced by environmental and genetic factors and/or exposure to treatment, resulting in a wide range of possible barriers to therapeutic efficacy and a range of different response–progression profiles, each with particular implications for subsequent treatment [13].

For instance, hyperprogression is the paradoxical occurrence of rapid clinical and radiographic deterioration that may occur shortly after initiating treatment. Definitions of hyperprogression vary within the literature, with some classifying tumor growth rate (TGR) according to tumor diameter, others by tumor volume, and some according to Response Evaluation Criteria in Solid Tumors (RECIST) evidence of progression within 2 months. In the context of NSCLC, hyperprogression is generally defined as a ≥2-fold increase in TGR in patients with disease progression between baseline and first assessment by RECIST criteria at 8 weeks. Although hyperprogression is well documented and is not necessarily specific to immunotherapy approaches, its etiology remains unclear. Possible explanations include oncogenic signaling

activation, upregulation of alternative immune checkpoints, or modulation of other protumor immune subsets. Predictive studies have failed to show any association between hyperprogressive disease and a range of disease characteristics (e.g. tumor burden, histologic subtype, number of metastatic sites) or therapeutic approaches (previous lines of chemotherapy or prior treatment), but age appears to be a risk factor. In one study, almost 20 % of patients older than 65 years developed hyperprogressive disease compared with only 5 % of those aged 65 years or younger ($p = 0.018$) [16,17].

Another rare progression–response profile is that of pseudoprogression, estimated to occur in approximately 2–6 % of patients treated with immunotherapy [11]. Pseudoprogression is characterized by a transient enlargement of the tumor (or metastatic sites) before regression in size, and is attributed to an initial inflammatory reaction that may resemble a tumor flare [16]. Immuno-histochemical evaluations suggest baseline tumor cells may increase in number in parallel with an inflammatory response consisting of activated cytotoxic lymphocytes (CD8 + T cells), TIA-1 (an apoptosis-promoting protein) and granzyme B (a protein necessary for the induction of apoptosis by cytotoxic T cells). In light of its transient nature, in order to differentiate pseudoprogression from true progression and necessary clinical action, new radiographic assessment protocols have been developed that mandate specific time intervals and application of an immunotherapy-specific RECIST classification system before evaluation of response to immunotherapy [16,17].

It should be noted that hyperprogression and pseudoprogression are rare events in patients who receive first-line chemo-immunotherapy; they are seen mainly in patients treated with pembrolizumab monotherapy. In cases of hyperprogression on pembrolizumab monotherapy, treatment with platinum-based doublet chemotherapy is likely; treatment with a bevacizumab-containing doublet could be considered, given the potential immunomodulatory role of bevacizumab, but as yet, there is no clinical data to robustly support this strategy given the rarity of hyperprogression.

A third form of progression has been identified as oligoprogression. Oligometastatic disease is an intermediate state between localized and widespread metastatic cancer, often defined in NSCLC as fewer than five sites of disease. The term usually describes patients with synchronous or metachronous metastatic disease at presentation, but recent genomic studies have revealed distinct clonal evolution at each site of metastatic disease, leading to the hypothesis that individual sites may develop treatment resistance or increased metastatic potential independent of the primary site of disease or even other metastatic sites [15]. This phenomenon is particularly associated with oncogene-addicted NSCLC. Results from the phase II SABR-COMET trial (NCT01446744) indicate that patients with oligoprogression limited to only a few sites of disease may warrant treatment with locally ablative therapies [18], and further data are awaited from trials such as NRG-LU-002 (NCT03137771).

3. Mechanisms of resistance to immunotherapy

In the absence of robust data to delineate the effect of approved immunotherapies on emergent tumor biology, the mechanisms underpinning different patterns of response–progression could potentially guide subsequent treatment selection in order to optimize outcomes for individual patients.

Licensed PD-1/PD-L1 immune checkpoint inhibitors function by preventing one mechanism of immune evasion available to the tumor. The immune checkpoint molecule PD-1 is expressed on the extracellular surface of natural killer T cells, B cells, dendritic cells, monocytes/macrophages, and CD4+ and CD8 + T cells. PD-L1 is present on immune cells, as well as on certain types of cancer and stromal cells. Interaction of PD-1 with PD-L1 inhibits the activation, proliferation, and survival of T cells [19]. The PD-1 receptor also interacts with programmed death ligand-2 (PD-L2), which is selectively expressed on certain types of macrophages and tolerogenic dendritic cells. Under physiological conditions, PD-1, PD-L1, and PD-L2 play important roles in regulating T-cell activation during peripheral tolerance: macrophages and tolerogenic dendritic cells prevent autoimmunity by inhibiting autoreactive T cells that may cause tissue damage [19]. Activation of the PD-1/PD-L1 pathway can result in the induction of T cell-mediated anergy, apoptosis, and 'exhaustion', initiating T-cell suppression. Solid tumors can 'hijack' the PD-1/PD-L1 axis, causing overexpression of PD-L1 and inducing immune suppression and evasion, thus inhibiting the attack of conventional cytotoxic CD8 + T cells and preventing tumor lysis. Inhibition of the PD-1/PD-L1 pathway, therefore, triggers tumor antigen recognition, proliferation, infiltration, and activation of cytotoxic CD8 + T cells, resulting in an antitumor immune response [19].

Successful antitumor immune response following immunotherapy with PD-1/PD-L1 immune checkpoint inhibitors, therefore, requires the reactivation and clonal proliferation of antigen-experienced T cells present in the tumor microenvironment (TME) [13,14]. Successful processing, presentation, and recognition of tumor-associated peptide antigens is required to generate tumor-specific CD8 + T cells with tumoricidal potential. The first signal for T-cell activation occurs when a unique T-cell receptor recognizes a major histocompatibility complex (MHC)-bound tumor antigen. Full T-cell activation is then triggered by the binding of the co-stimulatory CD28 receptor on T cells by B7 on the antigen-presenting cells. The resultant tumor-specific CD8 + T cells subsequently differentiate into effector T cells before undergoing clonal expansion and trafficking to the TME, where they kill tumor cells displaying the tumor-associated antigen on human leukocyte antigen (HLA).

For long-term immunologic memory, a subset of effector T cells must differentiate into effector memory T (T_{EM}) cells; these are maintained for life and respond to antigen re-challenge [14]. Disease progression (or 'failure' of immune checkpoint blockade), can result from defects in any of the steps that underpin this process, i.e. from insufficient generation of antitumor T cells, inadequate tumor-specific T-cell function, or impaired formation of T-cell memory [14]. Mechanistic defects that impair the effect of PD-1 blockade can arise from a number of tumor-intrinsic or -extrinsic factors, presenting either at the time of initial immunotherapy trial (primary/innate resistance) or after a period of initial response, resulting in progression (acquired resistance) (Fig. 1) [13,14].

3.1. Insufficient generation of antitumor T cells

Successful immunotherapy with immune checkpoint inhibitors requires the reactivation of T cells directed at tumor-specific mutant proteins. Conversely, a lack of suitable neoantigens and alterations in antigen processing (and/or presentation) is associated with impaired antitumor immune response.

Tumor-intrinsic mechanisms of immune evasion, therefore, include genetic and epigenetic alterations that influence neoantigen formation, presentation, and/or processing. They also include alterations in cellular signaling pathways that disrupt the action of cytotoxic T cells. Alterations in genes encoding components of the antigen processing and/or presentation apparatus (e.g. class I MHC, beta-2 microglobulin [β2M]) can also lead to resistance to immune checkpoint inhibitor therapy. Furthermore, downregulation of HLA class I molecules and loss of β2M expression may also be implicated: loss of β2M expression results in impaired cell-surface expression of MHC class I, which in turn impairs antigen presentation to cytotoxic T cells [14,20].

3.2. Inadequate tumor-specific T-cell function

A range of tumor-intrinsic and -extrinsic factors can also contribute to inadequate tumor-specific T-cell function and diminished clinical effect of PD-1/PD-L1 immune checkpoint inhibitors. These include PD-

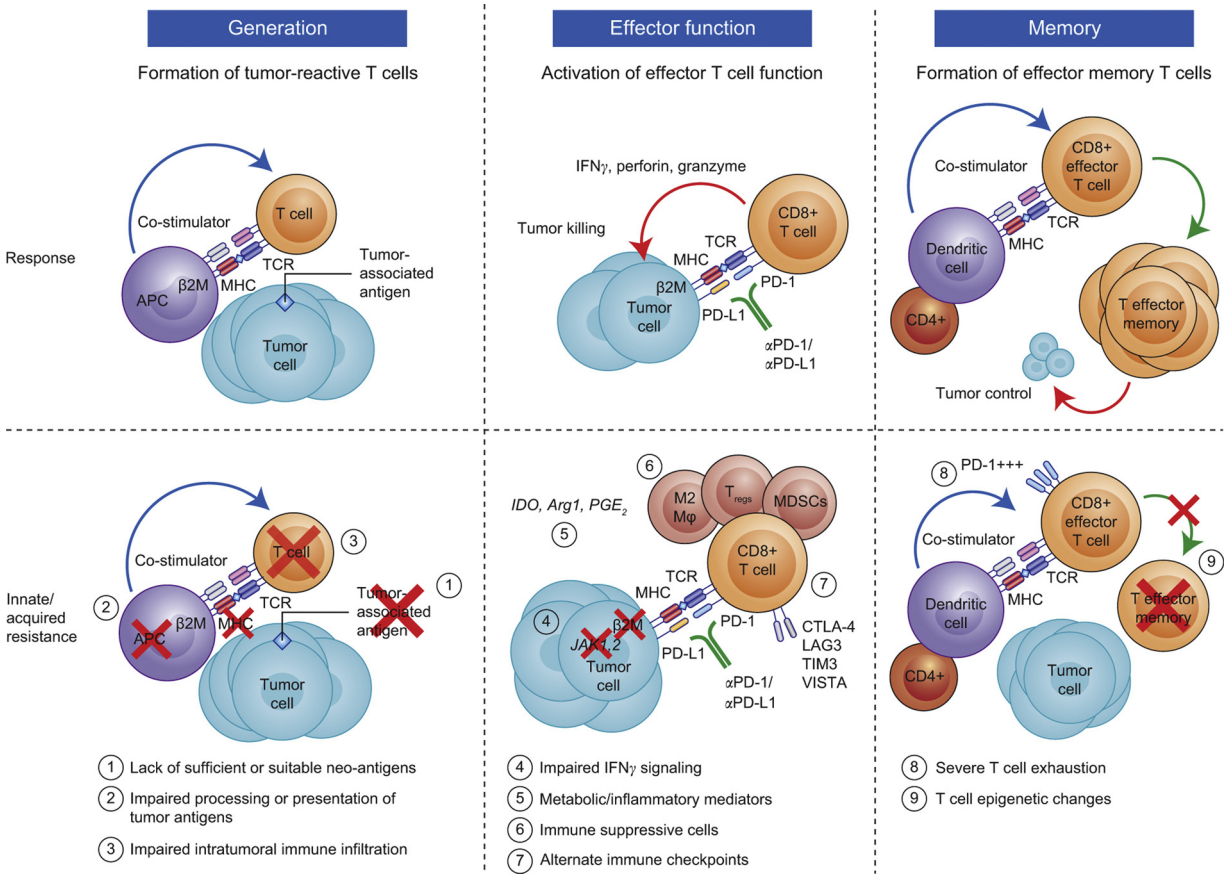

Fig. 1. Response and resistance to immune checkpoint inhibitor therapy.
Adapted from Jenkins et al. 2018 [14].
Upper panel: basic steps involved in generation of tumor-specific T cells, effector T-cell function, and formation of memory T cells.
Lower panel: putative mechanisms of innate and/or acquired resistance to immune checkpoint inhibitor therapy.
APC, antigen-presenting cells; αPD-1, anti-programmed cell death protein 1; αPD-L1, anti-programmed death ligand-1; Arg1, arginase 1; β2M, beta-2 microglobulin; CTLA-4, cytotoxic T-lymphocyte antigen 4; IDO, indolaimine-2, 3-deoxygenase; IFN, interferon; LAG3, lymphocyte activation gene 3; M2 Mϕ, type II macrophage; MDSC, myeloid-derived suppressor cell; MHC, major histocompatibility complex; PD-1, programmed cell death protein 1; PD-L1, programmed death ligand-1; PGE_2, prostaglandin E_2; TCR, T-cell receptor; TIM3, T-cell immunoglobulin and mucin domain-3; T_{reg}, regulatory T cell; VISTA, V-domain immunoglobulin suppressor of T-cell activation.

L1-independent mechanisms of immune escape, development of resistant mutations, T-cell exhaustion, and the development of an immunosuppressive TME.

For instance, a number of PD-L1-independent mechanisms of immune escape have been identified, such as immune suppressive cytokines, immune inhibitory metabolites, immune suppressive cells, and the over-expression of alternate immune checkpoints or co-inhibitory receptors (e.g. cytotoxic T-lymphocyte antigen 4 [CTLA-4], lymphocyte activation gene 3 [LAG3], T-cell immunoglobulin and mucin domain-3 [TIM3], and V-domain immunoglobulin-containing suppressor of T-cell activation [VISTA]) [13,14,21]. Targeting PD-1/PD-L1 in isolation, therefore, does not necessarily silence tumor pathogenesis mediated by one or a combination of these alternative pathways.

Resistance mutations can also arise that inhibit the clinical efficacy of ongoing immunotherapy. Whole exome sequencing of tumors from patients who developed resistance following initial clinical response to PD-1 blockade suggests that mutations in Janus kinases (JAK) 1 and 2 may be implicated [14]. Downregulating or mutating molecules involved in the interferon gamma signaling pathway (which goes through the interferon gamma receptor chains JAK1 and/or JAK2 and the signal transducer and activators of transcription [STATs]) could enable evasion of the effects of interferon gamma [13]. Preclinical data support the hypothesis that mutations or epigenetic silencing of molecules in the interferon receptor signaling pathway result in loss of the antitumor effects of interferon gamma, and analysis of tumors in patients who did not respond to anti-CTLA-4 therapy has revealed an enriched frequency of mutations in the interferon gamma pathway genes, interferon gamma receptor 1 and 2, JAK2, and interferon regulatory factor 1. Mutations in any of these genes resulting in inhibition of interferon gamma-related signaling could, therefore, result in primary resistance to anti-CTLA-4 therapy and lack of PD-L1 expression upon interferon gamma exposure [13].

Heterogeneity in PD-1 + and CD8 + populations also appears to be associated with differential response to PD-1 immune checkpoint inhibitor therapy and functional exhaustion of T cells. Indeed, partial exhaustion of PD-1 +, CTLA-4 +, and CD8 + infiltrating T cells has been shown to correlate with PD-1 response, and exhausted PD-1 + and CD8 + T cells display a distinct chromatin landscape compared with effector T cells and T_{EM} cells. These epigenetically distinct T cells appear to influence whether or not exhausted PD-1 + T cells can be reprogrammed to avoid terminal exhaustion and dysfunction [14].

3.3. Impaired formation of T-cell memory

While there is compelling evidence to support the ability of PD-1/PD-L1 immune checkpoint inhibitors to reinvigorate cytotoxic T lymphocytes and achieve long-term clinical benefit in some patients, this appears to be contingent upon effective T_{EM} formation. If T_{EM} formation

is impaired in any way, an initial clinical response may dissipate over time, leading to acquired resistance and potential disease progression.

Expansion of the intratumoral T_{EM} compartment has been demonstrated in response to PD-1 blockade, and to be positively associated with therapeutic response. Ongoing research aims to elucidate the mechanisms underlying T_{EM} expansion following PD-1 blockade, but distinct transcriptional programs associated with naïve, acute effector, memory, and exhausted T-cell states have been identified; there is emerging evidence that T-cell exhaustion is associated with epigenetic changes that appear to limit the durability of CD8 + T-cell function following PD-1 blockade [14].

3.4. Immunosuppression within the TME

The TME is also growing in recognition as another driver of acquired resistance to treatment with PD-1/PD-L1 immune checkpoint inhibitors, as immunosuppression within the TME has been shown to impair the efficacy of PD-L1 therapy [14,21].

An immunosuppressive TME is characterized by high levels of immune-suppressing cytokines and/or metabolites; by the recruitment of immune suppressive cells (e.g. myeloid-derived suppressor cells [MDSCs], and regulatory T cells [T_{regs}]); by mutations in key effector pathways; and by high levels of PD-L1 expression [14,21]. Preclinical models have shown an association between elevation of immune-suppressive cell types (including T_{regs}, MDSCs, Th2 CD4 + T cells, and M2-polarised tumor-associated macrophages) in the TME and diminished immune checkpoint inhibitor efficacy. These cell types promote an immunosuppressive TME that inhibits antitumor cytotoxic and Th1-directed T-cell activities, primarily through the release of cytokines, chemokines, and other soluble mediators [14].

Mutations associated with development of an immunosuppressive or 'cold' TME include *STK11/LKB1* and *KEAP1* (often found in conjunction with *RAS* mutations in NSCLC), which may result in primary resistance to immunotherapy [22–24]. For example, in the phase III MYSTIC study of durvalumab (anti-PD-L1) and tremelimumab (anti-CTLA-4), *STK11/LKB1* and *KEAP1* mutations were present in 16 % and 18 % of evaluable patients, respectively, and both were associated with poorer OS than in patients with wild-type variants; although there are currently no data to suggest *STK11/LKB1* or *KEAP1* mutations are predictive of a poorer response to immunotherapy compared with chemotherapy [25]. A retrospective analysis of patients with non-squamous NSCLC treated with first-line chemotherapy with or without pembrolizumab has shown that *STK11/LKB1* alterations define a subgroup of patients with inferior clinical outcomes and a lack of clinical benefit from addition of pembrolizumab [26]. The predictive value of *STK11/LKB1* and *KEAP1* mutations remains to be confirmed in a prospective clinical trial; current data suggest a strong prognostic potential, regardless of treatment [25].

4. Targeting immunotherapy resistance using anti-angiogenics

Considering progression on immunotherapy from a mechanistic perspective can help to identify therapeutic targets with the potential to delay or reverse resistance and prolong therapeutic effect. Among the most promising strategies under investigation are those targeting multiple mechanisms of immune escape using dual immune checkpoint inhibitor blockade (e.g. targeting both CTLA-4 and PD-1), and those using radiation (particularly stereotactic body radiation therapy) to prime immunity and enhance the effect of immunotherapy [13,14,27–29]. However, the approach with one of the strongest mechanistic foundations, and with immediate clinical relevance given the availability of licensed second-line agents, is the use of anti-angiogenics.

There is an increasing body of preclinical and clinical evidence to suggest that sustained angiogenesis and immunosuppression are interconnected processes with shared regulators [30–32]. Vascular abnormalities are a hallmark of many solid tumors and are known to facilitate immune evasion. The abnormalities stem from elevated levels of proangiogenic factors, such as vascular endothelial growth factor (VEGF) and angiopoietin 2 (ANG2). In addition to regulating angiogenesis, VEGF also plays a role in immunosuppression as abnormal vessels and impaired perfusion can restrict entry of cytotoxic drugs and immune cells from the circulation into tumors. To infiltrate the tumor and integrate into the TME, immune cells must enter the tumor blood vessels, adhere to the endothelium, and transmigrate across the vessel wall. The presence of angiogenic molecules such as VEGF within the TME can control the trafficking of immune cells to the tumor by altering the expression of adhesion molecules on endothelial cells (ECs) and immune cells [33].

VEGF also directly inhibits dendritic cell maturation and activation of antigen-specific T_{regs} and reduces immune cell–EC interactions in angiogenic vessels. Furthermore, there is a growing list of hematopoietic cell types (e.g. tumor-associated macrophages, MDSCs, TIE2+ monocytes, immature dendritic cells, and T_{regs}) that, when appropriately polarized, can promote both immunosuppression and angiogenesis through production of VEGF and other factors, such as basic fibroblast growth factor, chemokine (C-C-motif) ligand 2 and ANG2 [32]. Experimental studies have shown that depletion of these immunosuppressive cell types can enhance antitumor immune responses and has the potential to overcome innate resistance [14].

The discovery that VEGF was a key mediator of angiogenesis marked it out as a key therapeutic target, and led to the development of a number of agents with the potential to induce regression of angiogenic vessels and starve tumors of their blood supply and nutrients [33]. There are currently two anti-angiogenic agents licensed for use in metastatic NSCLC without oncogenic driver mutations in combination with docetaxel following progression on chemotherapy: nintedanib and ramucirumab [30,31].

In addition to its ability to suppress sprouting angiogenesis and delay tumor growth, anti-angiogenic activity can transiently normalize tumor vasculature and offer complementary immunomodulatory effects. Normalization of the tumor vasculature can improve blood perfusion and oxygenation, thereby enabling increased infiltration of immune effector cells and converting an immunosuppressive microenvironment to an immunosupportive environment (Fig. 2) [33–35].

This hypothesis is supported by preclinical evidence of an 'angio-immunogenic switch' mechanism whereby anti-angiogenics create a transient window for immune system detection and infiltration of anticancer therapies into the TME. In a model of immune-tolerant breast cancer, lower-doses (20 mg/kg or 10 mg/kg) of an anti-VEGFR2 antibody (DC101) have been shown to result in a more homogeneous distribution of functional blood vessels and improved tissue perfusion than full-dose DC101 (40 mg/kg). The lower-dose also enhanced the anticancer efficacy of vaccine therapy, reducing tumor volume and significantly slowing tumor growth compared with full-dose DC101. The lower-dose DC101 regimen reverted the immunosuppressive TME to an immunosupportive environment, with T-cell tumor infiltration inversely correlated with the DC101 dose [36].

Mechanistically, Wei et al. further demonstrate that the local immune landscape and PD-L1 heterogeneity may give rise to differing cancer severity hallmarks and clinical outcomes, predisposing some tumors to particular angiogenic and treatment response signatures. The authors report that nuclear factor kappa B signals elicited by macrophage inflammatory responses generate PD-L1+ cancer cells with aggressive survival capabilities, which support angiogenesis and have the ability to metastasize. Meanwhile, STAT1 signals triggered by activated T cells generate PD-L1+ cancer cells susceptive to apoptosis (Fig. 3) [37].

At the time of writing, there are no published trial data from prospective, randomized controlled trials evaluating the role of anti-angiogenic therapy following progression on immunotherapy (with/

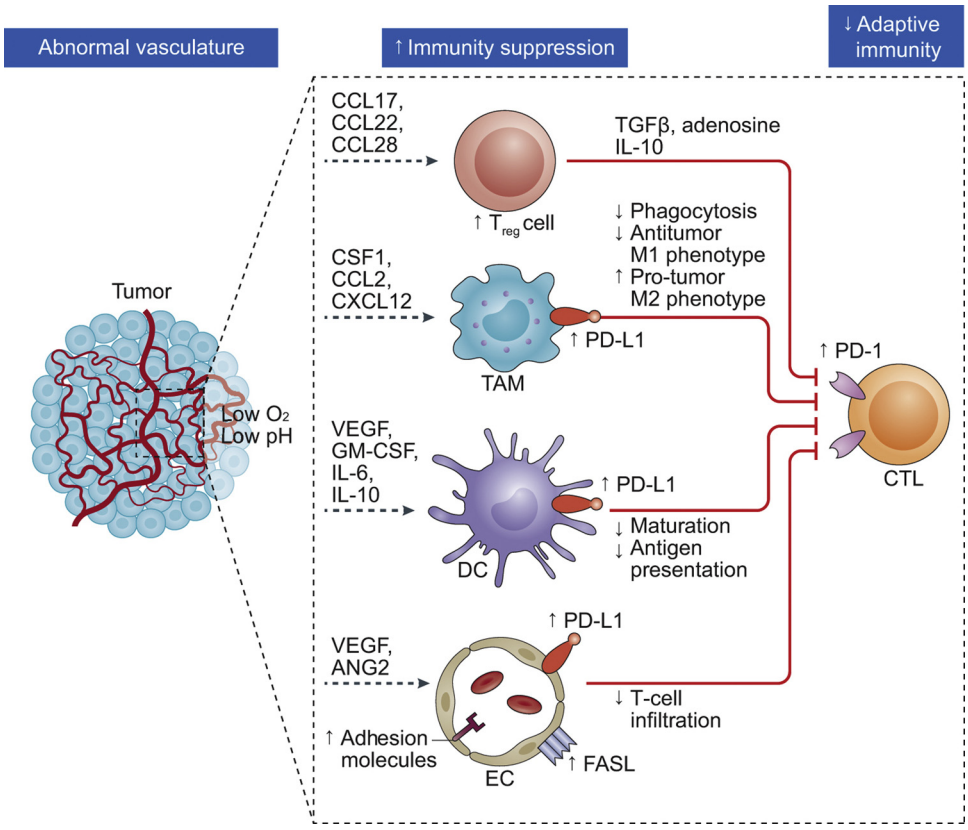

Fig. 2. Abnormal tumor vasculature contributes to immunosuppression in the TME. Adapted from Fukumura et al. 2018 [33]. Abnormalities in the tumor vasculature result in hypoxia and acidosis of the TME, which in turn contribute to immunosuppression via several mechanisms. These mechanisms include: increased accumulation, activation, and expansion of immunosuppressive T_{regs}; recruitment of inflammatory monocytes and TAMs; suppression of DC maturation, which results in impaired antigen presentation and activation of tumor-specific CTLs; and expansion of abnormal ECs with immunosuppressive phenotypes. Importantly, the PD-1/PD-L1 pathway is often activated in the TME as a mechanism to evade anticancer immune responses, with upregulation of PD-L1 expression on TAMs, DCs, and ECs, as well as on tumor cells. In addition, tumor-infiltrating CTLs typically upregulate PD-1, marking them as dysfunctional or 'exhausted' and limiting their cytotoxic potential against tumor cells. Overall, the consequence of aberrant tumor angiogenesis and vascular abnormality is an immunosuppressive TME.
ANG2, angiopoietin 2; CCL, C-C-motif chemokine ligand; CXCL12, C-X-C-motif chemokine ligand 12; CSF1, macrophage colony-stimulating factor 1; CTL, cytotoxic T lymphocyte; EC, endothelial cell; DC, dendritic cell; FASL, Fas ligand; GM-CSF, granulocyte–macrophage colony-stimulating factor; IL, interleukin; PD-1, programmed cell death protein 1; PD-L1, programmed death ligand-1; TAM, tumor-associated macrophage; TME, tumor microenvironment; TGFβ, transforming growth factor beta; T_{reg}, regulatory T cell; VEGF, vascular endothelial growth factor.

without chemotherapy). Prospective data will be generated by trials such as the planned trial of the combination of nab-paclitaxel and nintedanib or nab-paclitaxel and placebo in relapsed NSCLC adenocarcinoma (NCT03361319). In the meantime, however, published data are available from two real-world evaluations of nintedanib plus docetaxel following progression after chemotherapy followed by PD-1 blockade [38,39].

The first study involved a retrospective analysis of centers participating in the Spanish nintedanib named patient use program. Eligible patients (n = 11) received nintedanib plus docetaxel after progression on chemotherapy and immune checkpoint inhibitor therapy. Third-line treatment with nintedanib plus docetaxel was associated with an objective response rate (ORR) of 36 %, a disease control rate (DCR) of 82 %, and a median progression-free survival (PFS) of 3.2 months [38].

These findings are reinforced by the interim results from the non-interventional VARGADO study, involving patients (n = 40) who received nintedanib plus docetaxel following first-line chemotherapy and second-line immune checkpoint inhibitor therapy [39,40]. At the time of analysis (data cut-off: August 1, 2019), median duration of follow-up was 7.1 months for patients treated with nintedanib plus docetaxel. Median PFS was 7.2 months (95 % CI: 2.9–8.7). ORR and DCR data were available for 29 patients: partial response rate was 45 % and DCR was 86 %. Grade ≥3 treatment-emergent adverse events occurred in 43 % of patients; serious treatment-emergent adverse events occurred in 48 % of patients; and 30 % of patients discontinued due to treatment-emergent adverse events [40].

There are also complementary data for ramucirumab plus docetaxel following failure of nivolumab in metastatic NSCLC. Among patients (n = 20) included in a published retrospective analysis, ORR was 60 % and DCR was 90 %. Six patients had stable disease and two had progressive disease. Gastrointestinal adverse events were frequently observed in almost all (n = 19/20) patients [41].

Although small, these retrospective analyses provide initial evidence, in a real-world setting, demonstrating consistent clinical benefit with third-line anti-angiogenic therapies (in combination with docetaxel) after failure on a checkpoint inhibitor. Thus, the rational sequencing of anti-angiogenics after failure on immunotherapy may be a promising approach that warrants further investigation in future clinical trials.

Anti-angiogenics may also be combined with immunotherapy and chemotherapy. Recent data from the phase III IMPOWER150 trial offer proof of concept of the clinical relevance of the interplay between angiogenesis and immunosuppression. The trial investigated the combination of atezolizumab (A) plus anti-angiogenic bevacizumab (B) plus chemotherapy (C) versus AC and BC in chemotherapy-naïve patients with metastatic non-squamous NSCLC. Addition of atezolizumab to BC (i.e. ABC) resulted in a significant improvement in both PFS and OS, regardless of patients' PD-L1 expression or *EGFR* and *ALK* genetic alteration status. The authors proposed that the efficacy of atezolizumab may have been enhanced by the addition of bevacizumab and its ability to reverse VEGF-mediated immunosuppression [42]. This was particularly evident for the subset of patients with *EGFR*-mutant NSCLC, in which the bevacizumab-containing combination ABC resulted in an improved OS compared with the BC control [43]. As this survival advantage was not observed in the AC investigational arm, the results suggest a potential synergistic effect for bevacizumab and atezolizumab, at least in patients with *EGFR*-mutant NSCLC.

Ongoing or planned trials investigating anti-angiogenics in combination with immunotherapies in patients with NSCLC in a post-(chemo)-immunotherapy setting include: a trial of nivolumab, ipilimumab, and nintedanib in patients who develop resistance to immune checkpoint inhibitor therapy (as well as a separate treatment arm in newly diagnosed patients; NCT03377023); a trial of ramucirumab with atezolizumab in patients previously treated with an immune checkpoint

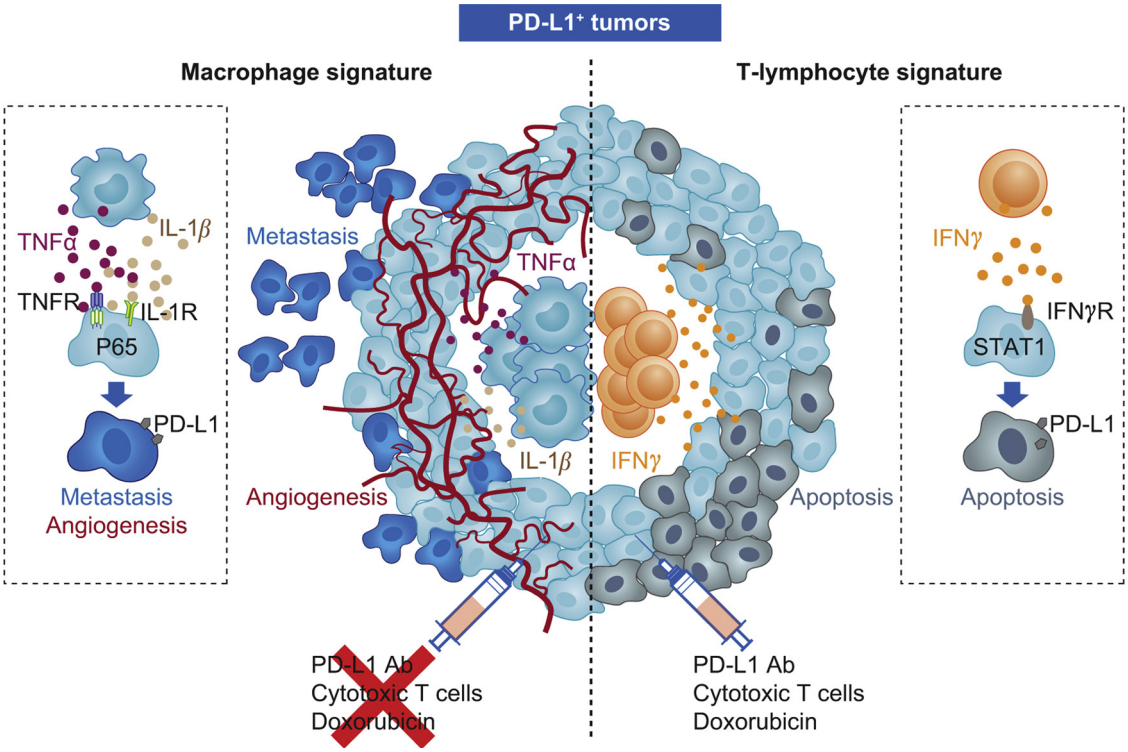

Fig. 3. Role of the local immune landscape on tumor PD-L1 heterogeneity and sensitivity to therapy.
Adapted from Wei et al. 2019 [37].
NF-κB signals elicited by macrophage inflammatory responses generate PD-L1 + cancer cells with survival, angiogenic, and metastatic capabilities. Meanwhile STAT1 signals triggered by activated T cells can induce susceptibility to apoptosis in PD-L1 + cancer cells.
Ab, antibody; IFN, interferon; IL, interleukin; NF-κB, nuclear factor kappa B; PD-L1, programmed death ligand-1; STAT, signal transducer and activator of transcription; TNF, tumor necrosis factor.

inhibitor, either alone or in combination (NCT03689855); and a trial of ramucirumab with nivolumab in patients who progressed after immunotherapy (either alone or in combination; the trial will also investigate immunotherapy-naïve patients; NCT03527108).

Irrespective of their future position within advanced NSCLC treatment protocols, optimizing the clinical benefit of anti-angiogenics will require consideration of the apparent dose- and time-dependent nature of their immunomodulatory effects [19,35].

5. Future perspectives

A recent Delphi consensus exercise conducted in Spain asked experts to consider the optimal selection of second- or later-line treatments following progression on first-line immunotherapy ± chemotherapy for advanced adenocarcinoma; in line with current ESMO guidelines, combined docetaxel and nintedanib was considered to be a valid option for patients progressing on prior lines of chemo-immunotherapy [2,44]. Nevertheless, prospective clinical data are essential to ensure that management decisions for NSCLC are supported by a robust evidence base. As the collective experience with first-line immunotherapy for metastatic NSCLC matures, the implications of different therapeutic approaches and their impact on tumor profile and subsequent treatment response will become clearer, helping clinicians navigate current therapeutic uncertainties. In parallel, targeted research efforts will be required, not only to provide proof-of-concept data for emerging therapeutic strategies, but also to facilitate evaluation of their long-term risk–benefit profiles and health economic implications.

Balancing therapeutic effect with tolerability will be particularly pertinent when assessing the suitability of combination approaches and treatment sequencing in elderly patients, in individuals with poorer PS and comorbidities, and in populations more representative of real-world case-loads. Of interest within this context is the open-label, phase IIb SENECA trial of nintedanib plus 3-weekly or weekly schedules of docetaxel. The weekly docetaxel schedule (33 mg/m^2 on days 1 and 8 of each 21-day cycle) was better tolerated than the 3-weekly schedule (75 mg/m^2), with no statistically significant difference in efficacy [45]. Furthermore, balancing health and quality of life outcomes with affordability will be non-trivial in light of the high economic impact that immunotherapies present to healthcare systems [46].

Central to achieving research efficiencies and the smooth translation of emerging knowledge into effective clinical approaches will be the development and use of a standard taxonomy for resistance classification and response–progression profiles. Ideally, related studies will also include paired re-biopsies at the time of progression, to enable better definition of the immunological stromal landscape, determination of biological correlates of resistance patterns, and new vulnerabilities that may benefit from the addition of an anti-angiogenic therapy.

6. Conclusions

Immunotherapy has markedly changed clinical algorithms for patients with non-oncogene-addicted, metastatic NSCLC. Yet, despite the meaningful successes in some patients, many ultimately relapse and the optimal choice of post-progression therapy remains to be determined. Until the availability of robust, prospective clinical trial data, joint decision making will be key to determining the risk–benefit profile of the currently available therapeutic options, and selecting the best option on an individual patient-by-patient basis [2].

In the meantime, consideration of the biological mechanisms of acquired tumor resistance to immunotherapy (and mapping these to the mechanisms of action of licensed therapeutic options) offers an interim

guide for assessing the validity of the available options. Within this context, targeting the TME appears to be the most promising approach to overcoming immunotherapy resistance using existing licensed agents. The intertwined regulation of VEGF signaling and immunosuppression in the TME clearly supports consideration of anti-angiogenic therapy to target immunosuppression in the TME and trigger an 'angio-immunogenic switch' back towards an immunosupportive environment [47].

Financial and competing interests disclosure

SP received personal fees from BMS, Roche, Takeda, AstraZeneca, Pfizer, MSD, EMD Serono, Guardant Health, AbbVie, Boehringer Ingelheim, Tesaro, OncLive and Medscape.

CG has received sponsorship or research funding and payment or other financial remuneration from AbbVie, AstraZeneca, Bayer, Boehringer Ingelheim, MSD, Pfizer and F. Hoffmann-La Roche, and payment or other financial remuneration from Lilly.

MR received fees for honoraria, consulting/advisory roles and speaker's bureau from Boehringer Ingelheim, F. Hoffmann-La Roche, Lilly, AstraZeneca, BMS, MSD, Merck, Novartis, Pfizer and Celgene.

SN received personal fees for advisor/speaker's bureau from AstraZeneca, Boehringer Ingelheim, BMS, Takeda, Pfizer, Roche, MSD, Eli Lilly and AbbVie.

JC and MG have no conflicts of interest.

DR is an employee of Boehringer Ingelheim International GmbH.

RK is an employee of Boehringer Ingelheim (Schweiz) GmbH and has a patent EP 2994125 issued.

Funding

Medical writing assistance was supported financially by Boehringer Ingelheim. SP acknowledges NHS funding to the Royal Marsden Hospital NIHR Biomedical Research Centre.

Transparency document

The Transparency document associated with this article can be found in the online version.

Acknowledgements

Medical writing assistance was provided by Alison Chisholm and Nirmal Jethwa on behalf of Syneos Health (London, UK) during the preparation of this review.

References

[1] International Agency for Research on Cancer, Cancer Today: Data visualization tools for exploring the global cancer burden in 2018, Cancer Today Data Vis. Tools Explor. Glob. Cancer Burd. 2018. (n.d.). http://gco.iarc.fr/today/home (accessed October 30, 2019).

[2] D. Planchard, S. Popat, K. Kerr, S. Novello, E.F. Smit, C. Faivre-Finn, T.S. Mok, M. Reck, P.E. Van Schil, M.D. Hellmann, S. Peters, Metastatic non-small cell lung cancer: ESMO clinical practice guidelines for diagnosis, treatment and follow-up, Ann. Oncol. 29 (Suppl 4) (2018) iv192–iv237, https://doi.org/10.1093/annonc/mdy275.

[3] National Comprehensive Cancer Network, Non-small Cell Lung Cancer (version 3), NCCN Clin. Pract. Guidel. Oncol., 2019.

[4] B. Dholaria, W. Hammond, A. Shreders, Y. Lou, Emerging therapeutic agents for lung cancer, J. Hematol. Oncol. 9 (1) (2016) 138, https://doi.org/10.1186/s13045-016-0365-z.

[5] G. Tsakonas, S. Ekman, Oncogene-addicted non-small cell lung cancer and immunotherapy, J. Thorac. Dis. 10 (Suppl 13) (2018) S1547–S1555, https://doi.org/10.21037/jtd.2018.01.82.

[6] L. Gandhi, D. Rodríguez-Abreu, S. Gadgeel, E. Esteban, E. Felip, F. De Angelis, M. Domine, P. Clingan, M.J. Hochmair, S.F. Powell, S.Y.-S. Cheng, H.G. Bischoff, N. Peled, F. Grossi, R.R. Jennens, M. Reck, R. Hui, E.B. Garon, M. Boyer, B. Rubio-Viqueira, S. Novello, T. Kurata, J.E. Gray, J. Vida, Z. Wei, J. Yang, H. Raftopoulos, M.C. Pietanza, M.C. Garassino, Pembrolizumab plus chemotherapy in metastatic non–small-cell lung cancer, N. Engl. J. Med. 378 (22) (2018) 2078–2092, https://doi.org/10.1056/NEJMoa1801005.

[7] S.M. Gadgeel, M.C. Garassino, E. Esteban, G. Speranza, E. Felip, M.J. Hochmair, S.F. Powell, S.Y. Cheng, H. Bischoff, N. Peled, R. Hui, M. Reck, T. Kurata, E.B. Garon, M.J. Boyer, J. Yang, M.C. Pietanza, D. Rodriguez-Abreu, KEYNOTE-189: Updated OS and progression after the next line of therapy (PFS2) with pembrolizumab (pembro) plus chemo with pemetrexed and platinum vs placebo plus chemo for metastatic nonsquamous NSCLC, J. Clin. Oncol. 37 (Suppl 15) (2019), https://doi.org/10.1200/JCO.2019.37.15_suppl.9013 9013–9013.

[8] J. Brahmer, H. Borghaei, S.S. Ramalingam, L. Horn, E. Holgado, A. Pluzanski, M.A. Burgio, M. Garassino, L.Q. Chow, S. Gettinger, L. Crino, D. Planchard, C. Butts, A. Drilon, J. Wojcik-Tomaszewska, G. Otterson, V. Hayreh, A. Li, J.R. Penrod, S.J. Antonia, Long-term survival outcomes with nivolumab (NIVO) in pts with previously treated advanced non-small cell lung cancer (NSCLC): impact of early disease control and response, Cancer Res. 79 (Suppl 13) (2019) CT195, https://doi.org/10.1158/1538-7445.AM2019-CT195.

[9] T.S.K. Mok, Y.-L. Wu, I. Kudaba, D.M. Kowalski, B.C. Cho, H.Z. Turna, G. Castro, V. Srimuninnimit, K.K. Laktionov, I. Bondarenko, K. Kubota, G.M. Lubiniecki, J. Zhang, D. Kush, G. Lopes, KEYNOTE-042 Investigators, Pembrolizumab versus chemotherapy for previously untreated, PD-L1-expressing, locally advanced or metastatic non-small-cell lung cancer (KEYNOTE-042): a randomised, open-label, controlled, phase 3 trial, Lancet Lond. Engl. 393 (10183) (2019) 1819–1830, https://doi.org/10.1016/S0140-6736(18)32409-7.

[10] L. Paz-Ares, A. Luft, D. Vicente, A. Tafreshi, M. Gümüş, J. Mazières, B. Hermes, F. Çay Şenler, T. Csőszi, A. Fülöp, J. Rodríguez-Cid, J. Wilson, S. Sugawara, T. Kato, K.H. Lee, Y. Cheng, S. Novello, B. Halmos, X. Li, G.M. Lubiniecki, B. Piperdi, D.M. Kowalski, Pembrolizumab plus chemotherapy for squamous non–small-cell lung cancer, N. Engl. J. Med. 379 (21) (2018) 2040–2051, https://doi.org/10.1056/NEJMoa1810865.

[11] M. Reck, R. Kaiser, A. Mellemgaard, J.-Y. Douillard, S. Orlov, M. Krzakowski, J. von Pawel, M. Gottfried, I. Bondarenko, M. Liao, C.-N. Gann, J. Barrueco, B. Gaschler-Markefski, S. Novello, Docetaxel plus nintedanib versus docetaxel plus placebo in patients with previously treated non-small-cell lung cancer (LUME-lung 1): a phase 3, double-blind, randomised controlled trial, Lancet Oncol. 15 (2) (2014) 143–155, https://doi.org/10.1016/S1470-2045(13)70586-2.

[12] E.B. Garon, T.-E. Ciuleanu, O. Arrieta, K. Prabhash, K.N. Syrigos, T. Goksel, K. Park, V. Gorbunova, R.D. Kowalyszyn, J. Pikiel, G. Czyzewicz, S.V. Orlov, C.R. Lewanski, M. Thomas, P. Bidoli, S. Dakhil, S. Gans, J.-H. Kim, A. Grigorescu, N. Karaseva, M. Reck, F. Cappuzzo, E. Alexandris, A. Sashegyi, S. Yurasov, M. Pérol, Ramucirumab plus docetaxel versus placebo plus docetaxel for second-line treatment of stage IV non-small-cell lung cancer after disease progression on platinum-based therapy (REVEL): a multicentre, double-blind, randomised phase 3 trial, Lancet 384 (9944) (2014) 665–673, https://doi.org/10.1016/S0140-6736(14)60845-X.

[13] P. Sharma, S. Hu-Lieskovan, J.A. Wargo, A. Ribas, Primary, adaptive, and acquired resistance to cancer immunotherapy, Cell 168 (4) (2017) 707–723, https://doi.org/10.1016/j.cell.2017.01.017.

[14] R.W. Jenkins, D.A. Barbie, K.T. Flaherty, Mechanisms of resistance to immune checkpoint inhibitors, Br. J. Cancer 118 (1) (2018) 9–16, https://doi.org/10.1038/bjc.2017.434.

[15] V. Tumati, P. Iyengar, The current state of oligometastatic and oligoprogressive non-small cell lung cancer, J. Thorac. Dis. 10 (Suppl 21) (2018) S2537–S2544, https://doi.org/10.21037/jtd.2018.07.19.

[16] J.S. Kurman, S.D. Murgu, Hyperprogressive disease in patients with non-small cell lung cancer on immunotherapy, J. Thorac. Dis. 10 (2) (2018) 1124–1128, https://doi.org/10.21037/jtd.2018.01.79.

[17] E. Borcoman, A. Nandikolla, G. Long, S. Goel, C. Le Tourneau, Patterns of response and progression to immunotherapy, Am. Soc. Clin. Oncol. Educ. Book (2018) 169–178, https://doi.org/10.1200/EDBK_200643.

[18] D.A. Palma, R. Olson, S. Harrow, S. Gaede, A.V. Louie, C. Haasbeek, L. Mulroy, M. Lock, G.B. Rodrigues, B.P. Yaremko, D. Schellenberg, B. Ahmad, G. Griffioen, S. Senthi, A. Swaminath, N. Kopek, M. Liu, K. Moore, S. Currie, G.S. Bauman, A. Warner, S. Senan, Stereotactic ablative radiotherapy versus standard of care palliative treatment in patients with oligometastatic cancers (SABR-COMET): a randomised, phase 2, open-label trial, Lancet 393 (10185) (2019) 2051–2058, https://doi.org/10.1016/S0140-6736(18)32487-5.

[19] R.R. Ramjiawan, A.W. Griffioen, D.G. Duda, Anti-angiogenesis for cancer revisited: Is there a role for combinations with immunotherapy? Angiogenesis 20 (2) (2017) 185–204, https://doi.org/10.1007/s10456-017-9552-y.

[20] J.M. Zaretsky, A. Garcia-Diaz, D.S. Shin, H. Escuin-Ordinas, W. Hugo, S. Hu-Lieskovan, D.Y. Torrejon, G. Abril-Rodriguez, S. Sandoval, L. Barthly, J. Saco, B. Homet Moreno, R. Mezzadra, B. Chmielowski, K. Ruchalski, I.P. Shintaku, P.J. Sanchez, C. Puig-Saus, G. Cherry, E. Seja, X. Kong, J. Pang, B. Berent-Maoz, B. Comin-Anduix, T.G. Graeber, P.C. Tumeh, T.N.M. Schumacher, R.S. Lo, A. Ribas, Mutations associated with acquired resistance to PD-1 blockade in melanoma, N. Engl. J. Med. 375 (9) (2016) 819–829, https://doi.org/10.1056/NEJMoa1604958.

[21] S.L. Topalian, C.G. Drake, D.M. Pardoll, Immune checkpoint blockade: a common denominator approach to cancer therapy, Cancer Cell 27 (4) (2015) 450–461, https://doi.org/10.1016/j.ccell.2015.03.001.

[22] K.C. Arbour, E. Jordan, H.R. Kim, J. Dienstag, H.A. Yu, F. Sanchez-Vega, P. Lito, M. Berger, D.B. Solit, M. Hellmann, M.G. Kris, C.M. Rudin, A. Ni, M. Arcila, M. Ladanyi, G.J. Riely, Effects of co-occurring genomic alterations on outcomes in patients with KRAS-mutant non–small cell lung cancer, Clin. Cancer Res. 24 (2) (2018) 334–340, https://doi.org/10.1158/1078-0432.CCR-17-1841.

[23] F. Skoulidis, M.E. Goldberg, D.M. Greenawalt, M.D. Hellmann, M.M. Awad, J.F. Gainor, A.B. Schrock, R.J. Hartmaier, S.E. Trabucco, L. Gay, S.M. Ali, J.A. Elvin, G. Singal, J.S. Ross, D. Fabrizio, P.M. Szabo, H. Chang, A. Sasson, S. Srinivasan,

S. Kirov, J. Szustakowski, P. Vitazka, R. Edwards, J.A. Bufill, N. Sharma, S.-H.I. Ou, N. Peled, D.R. Spigel, H. Rizvi, E.J. Aguilar, B.W. Carter, J. Erasmus, D.F. Halpenny, A.J. Plodkowski, N.M. Long, M. Nishino, W.L. Denning, A. Galan-Cobo, H. Hamdi, T. Hirz, P. Tong, J. Wang, J. Rodriguez-Canales, P.A. Villalobos, E.R. Parra, N. Kalhor, L.M. Sholl, J.L. Sauter, A.A. Jungbluth, M. Mino-Kenudson, R. Azimi, Y.Y. Elamin, J. Zhang, G.C. Leonardi, F. Jiang, K.-K. Wong, J.J. Lee, V.A. Papadimitrakopoulou, I.I. Wistuba, V.A. Miller, G.M. Frampton, J.D. Wolchok, A.T. Shaw, P.A. Jänne, P.J. Stephens, C.M. Rudin, W.J. Geese, L.A. Albacker, J.V. Heymach, STK11/LKB1 mutations and PD-1 inhibitor resistance in KRAS-mutant lung adenocarcinoma, Cancer Discov. 8 (7) (2018) 822–835, https://doi.org/10.1158/2159-8290.CD-18-0099.

[24] S.A. Best, D.P.D. Souza, A. Kersbergen, A.N. Policheni, S. Dayalan, D. Tull, V. Rathi, D.H. Gray, M.E. Ritchie, M.J. McConville, K.D. Sutherland, Synergy between the KEAP1/NRF2 and PI3K pathways drives non-small-cell lung cancer with an altered immune microenvironment, Cell Metab. 27 (4) (2018) 935–943, https://doi.org/10.1016/j.cmet.2018.02.006 e4.

[25] N. Rizvi, B.C. Cho, N. Reinmuth, K.H. Lee, A. Luft, M. Ahn, V. Papadimitrakopoulou, J. Heymach, U. Scheuring, B. Higgs, J. Ye, M. Kuziora, S. Wu, F. Liu, H. Si, S. Peters, OA04.07 Mutations associated with sensitivity or resistance to immunotherapy in mNSCLC: analysis from the MYSTIC trial, J. Thorac. Oncol. 14 (10) (2019) S217, https://doi.org/10.1016/j.jtho.2019.08.428.

[26] F. Skoulidis, K.C. Arbour, M.D. Hellmann, P.D. Patil, M.E. Marmarelis, M.M. Awad, J.C. Murray, J. Hellyer, J.F. Gainor, A. Dimou, C.M. Bestvina, C.A. Shu, J.W. Riess, C.M. Blakely, C.V. Pecot, L. Mezquita, F. Tabbò, M. Scheffler, V. Papadimitrakopoulou, J. Heymach, Association of STK11/LKB1 genomic alterations with lack of benefit from the addition of pembrolizumab to platinum doublet chemotherapy in non-squamous non-small cell lung cancer, J. Clin. Oncol. 37 (Suppl 15) (2019) 102, https://doi.org/10.1200/JCO.2019.37.15_suppl.102.

[27] M.D. Hellmann, T.-E. Ciuleanu, A. Pluzanski, J.S. Lee, G.A. Otterson, C. Audigier-Valette, E. Minenza, H. Linardou, S. Burgers, P. Salman, H. Borghaei, S.S. Ramalingam, J. Brahmer, M. Reck, K.J. O'Byrne, W.J. Geese, G. Green, H. Chang, J. Szustakowski, P. Bhagavatheeswaran, D. Healey, Y. Fu, F. Nathan, L. Paz-Ares, Nivolumab plus ipilimumab in lung cancer with a high tumor mutational burden, N. Engl. J. Med. 378 (22) (2018) 2093–2104, https://doi.org/10.1056/NEJMoa1801946.

[28] N. Shaverdian, A.E. Lisberg, K. Bornazyan, D. Veruttipong, J.W. Goldman, S.C. Formenti, E.B. Garon, P. Lee, Previous radiotherapy and the clinical activity and toxicity of pembrolizumab in the treatment of non-small-cell lung cancer: a secondary analysis of the KEYNOTE-001 phase 1 trial, Lancet Oncol. 18 (7) (2017) 895–903, https://doi.org/10.1016/S1470-2045(17)30380-7.

[29] A. Navarro-Martín, I. Galiana, M. Berenguer Frances, J. Cacicedo, R. Cañas Cortés, S. Comas Anton, S. Padrones Sánchez, S. Bolívar Cuevas, R. Parry, F. Guedea Edo, Preliminary study of the effect of stereotactic body radiotherapy (SBRT) on the immune system in lung cancer patients unfit for surgery: immunophenotyping analysis, Int. J. Mol. Sci. 19 (12) (2018) 3963, https://doi.org/10.3390/ijms19123963.

[30] Boehringer Ingelheim, Vargatef Summary of Product Characteristics, Vargatef SmPC. (n.d.). http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Product_Information/human/002569/WC500179970.pdf (accessed October 30, 2019).

[31] Eli Lilly, Cyramza Summary of Product Characteristics, Cyramza SmPC. (n.d.). https://www.ema.europa.eu/en/medicines/human/EPAR/cyramza (accessed October 30, 2019).

[32] L.F. Campesato, T. Merghoub, Antiangiogenic therapy and immune checkpoint blockade go hand in hand, Ann. Transl. Med. 5 (24) (2017), https://doi.org/10.21037/atm.2017.10.12 497.

[33] D. Fukumura, J. Kloepper, Z. Amoozgar, D.G. Duda, R.K. Jain, Enhancing cancer immunotherapy using antiangiogenics: opportunities and challenges, Nat. Rev. Clin. Oncol. 15 (5) (2018) 325–340, https://doi.org/10.1038/nrclinonc.2018.29.

[34] R.K. Jain, Normalizing tumor microenvironment to treat cancer: bench to bedside to biomarkers, J. Clin. Oncol. 31 (17) (2013) 2205–2218, https://doi.org/10.1200/JCO.2012.46.3653.

[35] R.K. Jain, Antiangiogenesis strategies revisited: from starving tumors to alleviating hypoxia, Cancer Cell 26 (5) (2014) 605–622, https://doi.org/10.1016/j.ccell.2014.10.006.

[36] Y. Huang, J. Yuan, E. Righi, W.S. Kamoun, M. Ancukiewicz, J. Nezivar, M. Santosuosso, J.D. Martin, M.R. Martin, F. Vianello, P. Leblanc, L.L. Munn, P. Huang, D.G. Duda, D. Fukumura, R.K. Jain, M.C. Poznansky, Vascular normalizing doses of antiangiogenic treatment reprogram the immunosuppressive tumor microenvironment and enhance immunotherapy, Proc. Natl. Acad. Sci. 109 (43) (2012) 17561–17566, https://doi.org/10.1073/pnas.1215397109.

[37] Y. Wei, Q. Zhao, Z. Gao, X.-M. Lao, W.-M. Lin, D.-P. Chen, M. Mu, C.-X. Huang, Z.-Y. Liu, B. Li, L. Zheng, D.-M. Kuang, The local immune landscape determines tumor PD-L1 heterogeneity and sensitivity to therapy, J. Clin. Invest. 129 (8) (2019) 3347–3360, https://doi.org/10.1172/JCI127726.

[38] J. Corral, M. Majem, D. Rodríguez-Abreu, E. Carcereny, Á.A. Cortes, M. Llorente, J.M. López Picazo, Y. García, M. Domine, M.P. López Criado, Efficacy of nintedanib and docetaxel in patients with advanced lung adenocarcinoma treated with first-line chemotherapy and second-line immunotherapy in the nintedanib NPU program, Clin. Transl. Oncol. 21 (9) (2019) 1270–1279, https://doi.org/10.1007/s12094-019-02053-7.

[39] C. Grohé, W. Gleiber, S. Haas, C. Losem, H. Mueller-Huesmann, M. Schulze, C. Franke, N. Basara, J. Atz, R. Kaiser, Nintedanib plus docetaxel after progression on immune checkpoint inhibitor therapy: insights from VARGADO, a prospective study in patients with lung adenocarcinoma, Future Oncol. 15 (23) (2019) 2699–2706, https://doi.org/10.2217/fon-2019-0262.

[40] C. Grohé, W. Gleiber, S. Kruger, H. Muller-Huesmann, M. Schulze, J. Atz, R. Kaiser, Efficacy and safety of nintedanib plus docetaxel in lung adenocarcinoma patients following treatment with immune checkpoint inhibitors: updated results of the ongoing non-interventional study VARGADO (NCT02392455), Ann. Oncol. 30 (suppl_11) (2019) xi16–xi32, https://doi.org/10.1093/annonc/mdz449.

[41] A. Shiono, K. Kaira, A. Mouri, O. Yamaguchi, K. Hashimoto, T. Uchida, Y. Miura, F. Nishihara, Y. Murayama, K. Kobayashi, H. Kagamu, Improved efficacy of ramucirumab plus docetaxel after nivolumab failure in previously treated non-small cell lung cancer patients, Thorac. Cancer 10 (4) (2019) 775–781, https://doi.org/10.1111/1759-7714.12998.

[42] M.A. Socinski, R.M. Jotte, F. Cappuzzo, F. Orlandi, D. Stroyakovskiy, N. Nogami, D. Rodríguez-Abreu, D. Moro-Sibilot, C.A. Thomas, F. Barlesi, G. Finley, C. Kelsch, A. Lee, S. Coleman, Y. Deng, Y. Shen, M. Kowanetz, A. Lopez-Chavez, A. Sandler, M. Reck, Atezolizumab for first-line treatment of metastatic nonsquamous NSCLC, N. Engl. J. Med. 378 (24) (2018) 2288–2301, https://doi.org/10.1056/NEJMoa1716948.

[43] M. Reck, R. Jotte, T.S.K. Mok, D.W.-T. Lim, F. Cappuzzo, F. Orlandi, D. Stroyakovskiy, N. Nogami, D. Rodríguez-Abreu, D. Moro-Sibilot, C.A. Thomas, F. Barlesi, G. Finley, M. Nishio, A. Lee, G. Shankar, W. Yu, M.A. Socinski, IMpower150: An exploratory analysis of efficacy outcomes in patients with EGFR mutations, Ann. Oncol. 30 (Suppl 2) (2019) ii38–ii68, https://doi.org/10.1093/annonc/mdz063.

[44] D. Isla, J. de Castro, R. García-Campelo, P. Lianes, E. Felip, P. Garrido, L. Paz-Ares, J.M. Trigo, Treatment options beyond immunotherapy in patients with wild-type lung adenocarcinoma: a Delphi consensus, Clin. Transl. Oncol. 22 (5) (2020) 759–771, https://doi.org/10.1007/s12094-019-02191-y.

[45] E. Capelletto, M.R. Migliorino, A. Morabito, R. Chiari, F. Grossi, M. Tiseo, F.D. Costanzo, A. Delmonte, G. Romano, D. Galetta, V. Scotti, V. Gregorc, S. Pisconti, G.L. Ceresoli, A. Del Conte, L. Ciuffreda, I. Colantonio, E. Bria, S. Ricciardi, A. Manzo, G. Metro, A.M. Morelli, R. Critelli, M.V. Pacchiana, I. Stura, G. Migliaretti, S. Novello, Final results of the SENECA (SEcond line NintEdanib in non-small cell lung CAncer) trial, Lung Cancer 134 (2019) 210–217, https://doi.org/10.1016/j.lungcan.2019.06.028.

[46] S. Thungappa, J. Ferri, C. Caglevic, F. Passiglia, L. Raez, C. Rolfo, Immune checkpoint inhibitors in lung cancer: the holy grail has not yet been found, ESMO Open 2 (1) (2017) e000162, https://doi.org/10.1136/esmoopen-2017-000162.

[47] C. Manegold, A.-M.C. Dingemans, J.E. Gray, K. Nakagawa, M. Nicolson, S. Peters, M. Reck, Y.-L. Wu, O.T. Brustugun, L. Crinò, E. Felip, D. Fennell, P. Garrido, R.M. Huber, A. Marabelle, M. Moniuszko, F. Mornex, S. Novello, M. Papotti, M. Pérol, E.F. Smit, K. Syrigos, J.P. van Meerbeeck, N. van Zandwijk, J. Chih-Hsin Yang, C. Zhou, E. Vokes, The potential of combined immunotherapy and anti-angiogenesis for the synergistic treatment of advanced NSCLC, J. Thorac. Oncol. 12 (2) (2017) 194–207, https://doi.org/10.1016/j.jtho.2016.10.003.