Is There an Optimal Intersection for Targeted and Immunotherapy Treatments for Melanoma?

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Abstract

Melanoma therapeutics have undergone massive changes with the approval of BRAF and MEK kinase inhibitors and immune-checkpoint blocking antibodies against CTLA-4 and PD-1. Targeted and immunotherapy have different strengths and weaknesses, but both are essential to clinical management of patients with advanced melanoma. Kinase inhibitors have rapid and high response rates, though their benefit is modest in terms of progression-free survival. Immunotherapy generally has lower response rates and can manifest atypical treatment kinetics. However, immunotherapy may offer a more robust potential for durable tumor control. Overall survival has been improved by both approaches, and next steps in clinical research will focus on how to combine these modalities. Early combination clinical trials have suggested that a cautious approach is appropriate when combining kinase inhibitors with immunotherapy. To further explore this, rigorous biomarker-driven clinical trials are essential. Beyond just melanoma, it seems likely that both combinations and sequenced approaches of targeted and immunotherapies will be required to harness the full potential of these approaches for treatment of cancer.

Keywords: BRAF, CTLA-4, MEK, melanoma, PD-1

Introduction

The clinical management of patients with *BRAF*-mutant advanced melanoma has undergone a dramatic transition over the past several years with the development of targeted kinase inhibitor approaches as well as immune-checkpoint blockade. Since 2011, 6 drugs have obtained FDA approval for the treatment of patients with advanced melanoma. These drugs include ipilimumab, the fully human monoclonal antibody against cytotoxic T-lymphocyte antigen 4 (CTLA-4); vemurafenib and dabrafenib, both serine-threonine kinase inhibitors targeting the mutant protein $BRAF^{V600}$; the MEK1/2 kinase inhibitor trametinib; and the anti-programmed death 1 (PD-1) antibody pembrolizumab. The first 4 of these drugs have been shown to improve the overall survival (OS) of patients with melanoma in phase 3 clinical trials,[1-5] and data for pembrolizumab are maturing.[6]

The PD-1 antibody nivolumab, approved in December 2014, also has shown an improvement in OS compared with dacarbazine chemotherapy in patients with *BRAF* wild-type melanoma,[7] and the combination of nivolumab with ipilimumab appears to improve response rates more than anti-PD-1 antibodies alone.[8] Additionally, the MEK1/2 inhibitor cobimetinib has improved OS when administered in combination with vemurafenib.[9] In considering these treatment options for a patient, it is clear that the response rate of kinase inhibitors targeting mutant *BRAF* is high; however, response duration is limited in most patients. Ipilimumab has a lower response rate and longer time to treatment benefit, but has been associated with approximately 20% survival at 3 years and potentially long-term survival thereafter in patients without disease progression at that point.[10] Anti-PD-1 antibodies can achieve substantial response rates (21%-40%)[6,7,11] with relatively rapid onset and durability of responses, while anti-PD-1 with anti-CTLA-4 may be even more robust.

Both targeted and immunotherapeutic treatments are important in the management of *BRAF*-mutant melanoma; however, the optimal sequence and combination of the available treatment agents is not clear. Because of the substantial antitumor activity in *BRAF*-mutant melanoma, inhibition of the mitogen-activated protein kinase (MAPK) pathway, through BRAF and MEK, may be particularly attractive to combine with immunotherapy. It has been hypothesized that antigen release through tumor cell death mediated by MAPK pathway inhibition may lead to increased antigen presentation or cross-presentation to tumor-specific T cells.[12] Inhibition of BRAF has also been found to increase the expression of melanoma differentiation antigens and induce infiltration of CD8+ T cells in posttreatment melanoma tumor samples.[13] Additionally, inhibition of BRAF and MEK in melanoma cells leads to increased tumor-specific T cell function, as well as dendritic cell function, in vitro.[14] Possible drawbacks of these kinase inhibitors when combined with CTLA-4 or PD-1/

programmed death ligand-1 (PD-L1) blockade may include the emergence of resistance and the potential for dampening of the immune response.[15]

Understanding the effects of kinase inhibitors on the immune response is thus essential for the development of rational combined targeted-immunotherapeutic drug regimens. Independent of immunotherapy, the combination of BRAF-MEK inhibition has predominately displaced single-agent BRAF inhibitor treatment, and it is therefore critical to delineate the impact of both a BRAF inhibitor and MEK inhibitor on the immune response.

Differential Immune Effects of Kinase Inhibitors

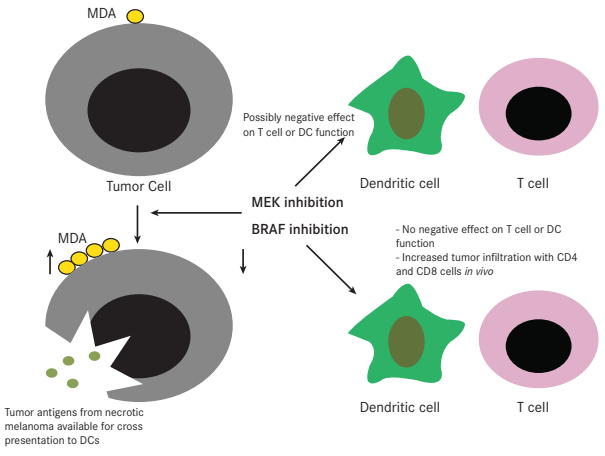
There is increasing evidence that kinase inhibitors exert effects on the immune system in addition to the tumor cells they are designed to target. The specific mechanisms by which this occurs are variable. Some examples include impact on the T cell receptor through Lck inhibition,[16] blockade of cytokine production through modulation of Src,[17] and suppression of myeloid-derived suppressor cells through KIT.[18] Multitarget kinase inhibitors (eg, imatinib) have been shown to have effects on broad immune cell populations such as CD4+ and CD8+ T cells,[19] natural killer (NK) cells,[20] and dendritic cells.[21] These effects can be positive or negative on the development of an antitumor response, with a productive example being observed in gastrointestinal stromal tumors, where imatinib has been shown to activate $CD8^+$ T cells and induce regulatory T cell apoptosis by reducing tumor-cell expression of indoleamine 2,3-dioxygenase.[22] This effect was augmented by CTLA-4 blockade.

Inhibition of $BRAF^{V600}$ by vemurafenib and dabrafenib appears to have relatively little effect on other kinases, and in vitro/in vivo modeling has suggested that BRAF inhibition improves T lymphocyte recognition of melanoma antigens and increases the numbers of $CD4^+$ and $CD8^+$ tumor infiltrating lymphocytes observed in melanoma tumors.[23] Other effects of BRAF inhibition include reduced expression of immunosuppressive cytokines,[24] increased activity of adoptively transferred T cells in mouse xenografts,[25] and decreased serum tumor necrosis factor-α.[26]

In contrast to $BRAF^{V600E}$ inhibition, MEK inhibition may have a negative impact on T-cell function. Notably, MEK inhibition at high doses has been shown to decrease proliferation and viability of T lymphocytes[13] (**Figure**). MEK inhibition also significantly decreases T-cell associated interferon-γ, whereas BRAF inhibitors do not. Further, MEK inhibition enhances the expression of forkhead box P3 (FoxP3)-positive T cells,[27] favoring T cell anergy and potentially contributing to an immunosuppressive tumor microenvironment. Finally, MEK inhibition, but not BRAF inhibition, negatively impacts dendritic cell cytokine secretion and decreases antigen presentation.[14]

A caveat to the impact on T cells by MEK inhibitors may be when they are used in combination with a BRAF inhibitor. In a small series of patient samples, no significant differences in quality of T cell infiltrate could be observed between patients receiving single-agent BRAF inhibitor versus BRAF-MEK inhibitor combination.[28] This may be due to the paradoxical activation of the MAPK pathway observed after administration of a BRAF inhibitor.[29] Whereas BRAF inhibitors block ERK signaling in *BRAF*-mutant melanoma, BRAF inhibitors have the opposite effect in *BRAF* wild-type immune cells, where they cause hyperactivation of ERK signaling. This can lead to enhancement of T-cell activation and may overcome the inhibitory effect of MEK inhibitors.[30] Thus, further research is needed to define the role of MEK inhibitors in combination with immunotherapy.

FIGURE. Effects of BRAF and MEK Inhibitors on Immune Cells



BRAF-mutant melanoma releases suppressive factors including cytokines and other molecules. Treatment with MEK or BRAF inhibitors increases the expression of melanoma differentiation antigens (MDA) and increases their presentation by antigen presenting cells such as dendritic cells (DCs). Whereas BRAF inhibition does not appear to have negative effects on immune cells, MEK inhibition may limit immune function and activation.

Clinical Experience

Preclinical data investigating the intersection between targeted and immunotherapies is developing and may guide future approaches. In the meantime, some lessons can be learned from studies already completed. Though initial reports are limited, the incidence of toxicity has been higher than expected. In renal cell carcinoma (RCC), the vascular endothelial growth factor receptor (VEGFR) inhibitor sunitinib was combined with tremelimumab, a CTLA-4 monoclonal antibody.[31] In this study, 9 of 28 patients with RCC experienced clinically significant toxicity,

predominantly transaminitis and acute renal failure. Preliminary data have been presented regarding the combination of the multitarget (including VEGFR) kinase inhibitors sunitinib and pazopanib with nivolumab, revealing improvement in response rates relative to historical controls but significant toxicity, with treatment being discontinued due to toxicity in 36% and 25% of patients, respectively.[32] These toxicities were also predominately hepatic and renal in nature.

In melanoma, the combination of kinase inhibitor and immune-checkpoint blockade has also been noted to manifest higher-than-expected toxicity. In the phase 1 study of vemurafenib and ipilimumab, 6 of 10 patients experienced dose-limiting immune-mediated hepatitis, leading to the combination being abandoned in clinical practice.[33] A similar phase 1 study of vemurafenib in combination with the anti-PD-L1 antibody MPDL3280A had to be modified to a staggered dosing approach for the 2 agents due to dose-limiting transaminitis.[34] The combination of dabrafenib and trametinib with ipilimumab was also discontinued due to the appearance of more severe colitis with bowel perforation. It did appear that dabrafenib without trametinib could be given safely with ipilimumab, though monitoring for immune-related hepatitis was recommended.[35] Based on these and other studies, hepatitis appears to be a common toxicity when combining targeted and immunotherapies.

The mechanisms of augmented immune-related adverse events seen with combination therapies are controversial. It is unknown whether these are related to kinase inhibitors or immune-checkpoint blockade. Although some of these combinations have shown encouraging clinical benefit, care will need to be given in developing combinations of kinase inhibitors with inhibitors of immune checkpoints, especially as anti-CTLA-4 with anti-PD-1 has also shown increased toxicity.[8]

Discussion

Remarkable advances with kinase inhibitors and immune checkpoint blockade have ushered in a new era in melanoma therapeutics. Combined BRAF and MEK inhibition yields rapid responses in most patients; however, the duration of benefit remains modest. Immune checkpoint blockade by ipilimumab, and possibly anti-PD1 antibodies, may offer a greater potential for durable disease control, though the response rates are lower and atypical treatment responses can be difficult to manage in clinic. In this context, there is hope that synergies between these fundamentally different approaches can be identified and translated into improved antitumor activity. Based on the experience of combination approaches to date, cautious and prudent evaluation will be necessary given the documented increased toxicity in early studies. This will be especially important as further combinations come into investigation such as PD-1 blockade with other immunomodulators (cytokines or other immune checkpoint inhibitors), VEGF inhibition, and chemotherapy.

In clinical practice, the appropriate choice of frontline therapy is guided by limited evidence. Many experts in the field suggest that consideration of immunotherapy initially is most prudent because, if the immune response is induced, it may be that no further therapy is needed. This is in contrast with targeted therapy, where treatment is open-ended and will almost always eventually fail. Some have also proposed that patients treated with targeted therapy initially have poorer outcomes with immunotherapy as second-line treatment, though data supporting this are preliminary.[36]

Moving forward, there are several studies evaluating concomitant or sequential administration of kinase inhibitors and immune checkpoint blockade. Examples include a study of BRAF-MEK inhibition with an anti-PD-1 antibody (dabrafenib, trametinib, pembrolizumab) in *BRAF*-mutant melanoma (ClinicalTrials.gov Identifier: NCT02130466), a study of the MEK inhibitor cobimetinib with the anti-PD-L1 antibody MPDL3280A in *NRAS*-mutant melanoma (ClinicalTrials.gov Identifier: NCT01988896), as well as sequential approaches of initial BRAF plus MEK (dabrafenib-trametinib) combination as compared with initial anti-CTLA-4 plus anti-PD-1 (ipilimumab-nivolumab) combination with eventual crossover to the alternate approach (ClinicalTrials.gov Identifier: NCT02224781).

Drug development in molecular and immunotherapy has set a new bar in advanced melanoma, and this paradigm is likely to become relevant to all cancers in the near future. The best approach in combining these therapies is an open question that urgently needs to be answered. As research moves forward, incorporation of the underlying tumor genotype will likely need to be taken into account given the emerging data that mutation in *NRAS* may increase response to immunotherapy.[37] Optimal sequencing and dosing of targeted and immunotherapy will only be elucidated in clinical trials, and the development of therapeutic biomarkers will be critical. It seems likely that both combination and sequential approaches of kinase inhibitors with immunotherapy will be required depending on the molecular biology of the patient's tumor (*BRAF* vs *NRAS* vs other) and the particular immunotherapy under study.

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