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REVIEW ARTICLE

Advances in the Systemic Treatment of Metastatic Melanoma

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ABSTRACT: Prior to 2011, the only commercially available agents commonly used to treat metastatic melanoma—including dacarbazine([Drug information on dacarbazine](#)), temozolomide([Drug information on temozolomide](#)) (Temodar), fotemustine, carboplatin([Drug information on carboplatin](#)), paclitaxel([Drug information on paclitaxel](#)), and interleukin-2—demonstrated limited efficacy, and no study involving these agents had shown an improvement in overall survival. The standard of care for the treatment of metastatic melanoma was radically changed by the subsequent approval of two agents, ipilimumab (Yervoy) and vemurafenib (Zelboraf), both of which improved survival in randomized phase III trials. Within the relatively short time that ipilimumab and vemurafenib have been commercially available, phase II data for the investigational agents nivolumab and MK-3475, for the combination of dabrafenib and trametinib, and for adoptive cell therapy strongly suggest even further improvements in treatment outcomes. Within this rich context of effective agents, the challenge for clinicians and investigators will be to develop predictive biomarkers of response, the optimal sequence of therapy for individual patients, and effective combinations. An additional challenge will be to find the appropriate venue and populations to test promising new agents arising from substantial advances in our understanding of molecular alterations in melanoma cells, of mechanisms of resistance to current agents, and of tumor-host immune interactions.

Introduction

Before 2011, no systemic treatment for unresectable locally advanced stage III or stage IV melanoma had been consistently proven to increase median survival, and no large studies had compared existing treatments to best supportive care. In large controlled randomized trials, the median survival was consistently in the range of 8 to 11 months.[1-3] High-dose interleukin-2 (IL-2) was approved for the treatment of metastatic melanoma based on durable tumor remissions in approximately 5% of patients, but it can only be administered to those with excellent performance status and normal organ function, and to date it has not been compared with any other standard treatment in a randomized trial.[4]

TABLE

Current advances in the treatment of advanced disease stem from the identification of two specific driver mutations in subsets of melanoma, *BRAF* and *C-KIT*, and from advances in our understanding of mechanisms that control T-lymphocyte activation, proliferation, and function, specifically the immune regulatory checkpoints (Table). Controlled clinical trials of vemurafenib (Zelboraf), which potently inhibits signaling from mutant BRAF, and of ipilimumab (Yervoy), which blocks the immune checkpoint cytotoxic T-lymphocyte antigen 4 (CTLA-4), demonstrated meaningful improvements in median survival.[5,6] Ipilimumab was also shown to produce a durable survival benefit in approximately 10% of patients. As a consequence, both vemurafenib and ipilimumab were approved by the US Food and Drug Administration (FDA) in 2011. Results of clinical trials of monoclonal antibodies designed to block another immune checkpoint, programmed death 1 (PD-1), or its ligand, and of combined inhibitors of mutant BRAF and MEK, suggest even further improvements in outcome for subsets of patients. Additional treatment gains may be achieved over the next 5 to 10 years through the combination of active agents, the introduction of new agents against novel molecular and immune targets, and improvements in technology that will increase the feasibility of adoptive cellular therapy outside of a few highly specialized treatment centers.

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graph TD
    A[T lymphocyte activation] --> B[Upregulation of cell surface co-inhibitory receptors on T lymphocytes]
    B --> C[Binding of co-inhibitory receptors to ligands on antigen-presenting cells or tumor cells]
    C --> D[Suppression of lymphocyte activation, proliferation, and/or function]
    D --> E[Blockade of co-inhibitory receptors or their ligands by antibodies]
    E --> F[Enhanced lymphocyte response and function]
    F --> G[Antitumor activity]
  
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Immune-Based Therapies

Ipilimumab is a human immunoglobulin G1 (IgG1) monoclonal antibody that blocks cytotoxic CTLA-4, a coinhibitory receptor that regulates T-cell activation and the function of T-regulatory cells (Figure). Approval followed presentation of results from a phase III trial that compared ipilimumab, 3 mg/kg every 3 weeks for 4 doses, to ipilimumab in combination with a gp100 peptide vaccine, or to the gp100 vaccine alone in patients who had received at least one prior treatment for advanced disease.[6] Although the objective response rate for ipilimumab in both arms combined was only 7%, median survival for patients receiving ipilimumab in either of the two arms was increased to 10 months, compared with 6.4 months in the vaccine-alone arm. Survival rates at 1 and 2 years were also improved for the ipilimumab arms, from 25% to 44%–46%, and

from 14% to 22%–24%, respectively. Recent long-term follow-up from earlier phase II trials of ipilimumab have shown that survival rates remain nearly flat from 3 to 5 years, indicating a long-term benefit for a subset of patients.[7] A second phase III trial was conducted in previously untreated patients, comparing ipilimumab at a dose of 10 mg/kg administered with dacarbazine to dacarbazine/placebo.[8] Although median survival in the ipilimumab plus dacarbazine arm was increased to 11.2 months from 9.1 months, the contribution of dacarbazine to the activity of ipilimumab remains unclear.

There are several unique features of ipilimumab treatment that have been described extensively in prior publications, including the induction of autoimmune/inflammatory adverse events and clinical response in small brain metastases in a subset of patients.[9] Several patterns of systemic tumor response have been observed, including mixed responses, disease progression followed by regression, and prolonged disease stabilization that appears to be associated with patient benefit.[10] The unique patterns of response led to the development of new criteria for assessing clinical response to immune therapy agents. Data from the initial phase III study of ipilimumab also demonstrated that patients treated with ipilimumab or ipilimumab plus vaccine, whose disease progressed after they had achieved stable disease or tumor response at 24 weeks, could achieve a second response or prolonged stable disease with a second induction course of ipilimumab.[11] Indeed, among 31 patients eligible for retreatment, objective response or stable disease of at least 24 weeks was observed in 68%. In contrast, the effect of administering maintenance ipilimumab, for example every 12 weeks, remains unclear.

Several combinations of ipilimumab with other agents may further increase activity and improve outcomes. Promising data, including increased overall response rate, progression-free survival, or complete response rate compared with results in prior trials, were presented for ipilimumab in combination with [bevacizumab](#) ([Drug information on bevacizumab](#)) (Avastin), ipilimumab in combination with high-dose IL-2, and tremelimumab (another anti-CTLA-4 antibody) in combination with interferon alfa.[12-14] A reliable predictive biomarker for response to ipilimumab has not yet been identified.[15-18]

Inhibitors of programmed death 1 (PD-1) or its ligand (PD-L1)

PD-1 is an inhibitory receptor that is upregulated on activated lymphocytes. PD-1 has two known ligands, PD-L1 (also called B7-H1) and PD-L2 (B7-DC), which can be expressed on tumor and stromal cells; PD-L1 expression can be induced by cytokines produced by tumor-infiltrating lymphocytes.[19-21] Several agents targeting either PD-1 or PD-L1 are being developed. In a phase I/II study of nivolumab (BMS-936558, MDX-1106), a human IgG4 monoclonal antibody that blocks PD-1, an overall objective response rate of 31% was observed among 106 evaluable patients with previously treated advanced melanoma.[22,23] An ongoing response was seen in 16 of 23 patients with objective response who were followed at least 6 months from onset of treatment. A similarly high objective response rate of 47% was observed among 83 patients with advanced melanoma who were treated with MK-3475, another antagonist antibody of PD-1.[24] Among the 25 patients in this group who had previously been treated with ipilimumab, MK-3475 produced an objective response rate of 40%. Overall, several complete responses were observed, and most patients were continuing in response with a minimum follow-up of 16 weeks. In a multitumor phase I trial of the anti-PD-L1 antibody BMS-936559, 9 of 52 melanoma patients (17%) achieved a complete or partial response.[25]

Toxicities associated with blockade of the PD-1 pathway have been similar in spectrum but less frequent and less severe than those seen with ipilimumab.[22] Grade 3 or 4 adverse events were observed in only 14% of patients treated with nivolumab and in 9% treated with the anti-PD-L1 agent BMS-936559. Pneumonitis was observed in 3% of patients treated with nivolumab and was fatal in 1%, leading to implementation of early detection and management algorithms in an attempt to reduce life-threatening reactions.

In the nivolumab phase I trial, a strong association was discovered between expression of PD-L1 in pretreatment tumor samples, defined as expression on 5% or more of tumor cells, and response to therapy.[22] Additional data will be required to confirm this association in melanoma. Studies conducted by Taube et al demonstrate that metastatic melanoma lesions that express PD-L1 are almost always associated

with the presence of tumor-infiltrating lymphocytes (TIL), while those metastatic lesions without PD-L1 expression generally have no TIL.[26] Increasing the activity of PD-1 blockade may require different approaches in the two subsets of tumors—for example, combining PD-1 blockade with other antagonists of lymphocyte functional suppression in PD-L1/TIL-positive tumors, and combining PD-1 blockade with agents that drive lymphocyte infiltration into tumors for those that are PD-L1/TIL-negative.

Adoptive cell therapy (ACT)

Existing immune therapies attempt to induce or expand tumor antigen-specific immune responses in vivo. An alternate approach is to isolate tumor antigen-specific T cells from the patient, either from peripheral blood or a resected tumor, and expand the cells ex vivo before reinfusing the cells back into the patient. Early studies of ACT in the late 1980s and early 1990s produced limited activity, believed to be a result of the limited persistence of the lymphocytes after adoptive transfer.[27,28] Preclinical models demonstrated that persistence of the cells in vivo after adoptive transfer could be increased if the host was preconditioned with lymphoablating chemotherapy and/or whole-body radiation.[29] Subsequent studies of lymphoablation, followed by transfer of TIL in combination with systemic administration of IL-2, demonstrated high response rates—in the range of 50%.[30-32] In the largest study published to date, approximately 20% of patients achieved durable complete remissions. Responses were observed in patients whose disease was progressing on anti-CTLA-4 therapy, and in a subsequent trial, we are aware of a patient responding after exposure to anti-PD-1 therapy, suggesting that ACT provides an antitumor effect that is non-cross-resistant to the checkpoint inhibitors.

Currently, ACT is applicable to only a select subset of patients who have good performance status and normal organ function, resectable tumors from which cells can be isolated and expanded, ability to travel to one of a few specialized centers studying ACT, and ability to maintain their performance while waiting for cells to expand in vitro for 3 to 6 weeks. Various technological advances may allow export of the technology to multiple centers and increase access to more patients—for example, by reducing the generation time and cost of expanding lymphocytes ex vivo. Better selection of antigen-specific T cells from resected tumors, improved expansion techniques, identification of populations with the greatest potential for in vivo activity, and improved approaches to the support of cell expansion and function after adoptive transfer (perhaps by concurrent administration of other cytokines and checkpoint inhibitors) may produce greater efficacy. Several trials have been conducted using peripheral blood lymphocytes that were genetically engineered ex vivo to express either a tumor-specific T-cell receptor or a chimeric antigen receptor (CAR).[33-36] CARs combine the signal-activating machinery of a T cell and the antigen binding site of a monoclonal antibody. By engineering peripheral blood lymphocytes to confer tumor antigen specificity, the costly and labor-intensive process of harvesting cells from tumors, and the concomitant delay in treatment, could possibly be avoided. Moreover, introducing tumor antigen-specific receptors to peripheral blood lymphocytes may extend therapy options to a larger group of patients. Some of the attempts to administer T cells transfected with CARs or specific T-cell receptors have been associated with unexpected toxicity, and overall response rates are currently lower than those reported with expanded TIL, but advances in the technology can be expected over time.[33]

Small-Molecule Antagonists of Tumor-Specific Mutations

Melanomas harboring BRAF mutations

The RAS-RAF mitogen-activated protein kinase (MAPK) intracellular signaling cascade has been shown to be critical for malignant behavior in the majority of melanomas. It directly impacts several cellular processes, including cell survival, differentiation, and proliferation. Somatic *BRAF* missense mutations present in approximately 40% to 60% of melanoma patients produce elevated kinase activity and activation of the MAPK pathway independent of upstream activation by RAS.[37] Mutations in *BRAF* are more common in cutaneous melanomas and are significantly less frequent in tumors in sun-shielded areas, such as mucosal or acral-lentiginous melanomas (0 to 9%, and approximately 15% to 23%, respectively).[38-41] Mutations in

BRAF are not found in uveal melanomas.[42,43] Approximately 80% to 90% of *BRAF* mutations are V600E, and 10% to 20% are V600K. Other rare mutations have been noted in the literature, some of which may be less responsive to the selective mutant *BRAF* inhibitors.[44]

Vemurafenib is a small-molecule potent inhibitor of mutant *BRAF*. [45] In assays conducted in vitro, it has little effect on melanoma cells with wild-type *BRAF* at concentrations that markedly inhibit the growth of cells carrying a mutation in *BRAF* V600E or V600K. Phase I and II studies demonstrated rapid antitumor activity in the majority of patients carrying a tumor with the *BRAF* V600E mutation. In a phase III trial in patients with *BRAF* V600E mutations, objective responses were observed in 48% of those who received vemurafenib and in 5% of those treated with dacarbazine.[5,46] The median progression-free survival in the vemurafenib arm was 6.9 months, compared with 1.6 months for dacarbazine. Vemurafenib increased median survival from 9.7 to 13.6 months despite eventual crossover from dacarbazine to vemurafenib.[5,46] Based on these remarkable data, vemurafenib was approved by the FDA in 2011. The most common side effects seen with vemurafenib were arthralgia, rash, fatigue, alopecia, keratoacanthoma, squamous cell carcinoma, photosensitivity, nausea, and diarrhea. Adverse effects necessitated a dose reduction in 38% of patients in the trial. Dabrafenib, another relatively selective inhibitor of mutated *BRAF*, was also compared to dacarbazine and produced an increase in median progression-free survival from 2.7 months to 5.1 months and an increase in the objective response rate from 7% to 50%—results similar to those seen with vemurafenib.[47] The toxicity profile of dabrafenib was also similar to that of vemurafenib, although pyrexia was noted more frequently.

Despite the impressive activity of vemurafenib and dabrafenib, current data indicate that most patients treated with either of these agents will develop progressive disease, and responses are generally not maintained when the drug is stopped.[48] Treatment can be continued after limited progression in some patients with probable additional benefit. Several mechanisms of resistance have been identified, some involving reactivation of signaling through downstream MEK and persistent phosphorylation of ERK.[49-53] In addition, in normal cells, vemurafenib and dabrafenib can activate MEK signaling through upstream activation of C-RAF, which is the cause of the secondary cutaneous squamous cell carcinomas observed in patients treated with these agents.[54-56] The identified mechanisms of tumor resistance and the development of secondary skin cancers suggested that the combined inhibition of mutant *BRAF* and MEK would produce improved antitumor effects and might reduce the skin-related toxicities seen with the *BRAF* inhibitors.

Trametinib is a small molecule that binds to and potentially inhibits MEK1 and MEK2.[57] A phase III trial compared trametinib to standard chemotherapy (dacarbazine or paclitaxel) in patients whose tumors contained a *BRAF* mutation. Median progression-free survival was improved from 1.5 to 4.8 months and overall survival at 6 months was increased from 67% to 81%. Crossover was allowed once patients had progressed on chemotherapy. Rash, diarrhea, and peripheral edema were the most common side effects.[58] Overall activity for the MEK inhibitor appeared to be less than for the mutant *BRAF* inhibitors in the same patient population. Trametinib was subsequently shown to have minimal activity and produced no objective responses in patients whose disease progressed on a *BRAF* inhibitor.[59]

In a phase I trial of dabrafenib combined with trametinib, full doses of both agents could be given together safely.[60] The combination was associated with a greater incidence of pyrexia, sometimes requiring concurrent administration of corticosteroids, but a lower incidence of cutaneous toxicities, including the development of cutaneous squamous cell carcinomas. Subsequently, 162 patients were randomly assigned to receive dabrafenib, 150 mg orally twice daily alone, or dabrafenib in combination with trametinib at either 1 or 2 mg orally daily. Median progression-free survival for the 150/2 combination group was 9.4 months, compared with 5.8 months in the dabrafenib-alone arm. The overall objective response rate was also higher in the 150/2 combination group, 76% vs 54% with dabrafenib alone ($P = .03$). Long-term follow-up data are not yet available to determine the effects on overall survival and on duration of responses; however, the results suggest that the combination will become the treatment of choice for targeting *BRAF* mutations in patients with metastatic melanoma.

Melanomas that harbor C-KIT mutations

C-KIT is a member of the receptor tyrosine kinase family of proteins. Activation of the intracellular signaling cascade by the endogenous ligand, stem cell factor, is involved in several cellular processes, including proliferation and inhibition of apoptosis.[61] *C-KIT* mutations are found in approximately 20% (range, 6% to 39%) of mucosal and acral-lentiginous melanomas, rarely in conjunctival melanomas, and in approximately 15% of melanomas arising from chronically sun-damaged skin.[62-67] Inhibitors of C-KIT tyrosine kinase, such as [imatinib\(Drug information on imatinib\)](#) (Gleevec), [dasatinib\(Drug information on dasatinib\)](#) (Sprycel), [sorafenib\(Drug information on sorafenib\)](#) (Nexavar), sunitinib (Sutent), and [nilotinib\(Drug information on nilotinib\)](#) (Tasigna) have been studied in melanoma patients. Trials in patients overexpressing *C-KIT* by immunohistochemistry demonstrated minimal activity.[68-71] Subsequently, several case reports and a few limited series provided evidence for the therapeutic activity of various C-KIT inhibitors in patients with tumors containing an activating *C-KIT* mutation.[65,72,73] In the largest treatment study reported in the literature, 21 patients with *C-KIT* mutations were treated with imatinib, and six objective responses were observed, including two complete responses. All responses occurred in patients with L576P or K642E mutations.[66]

Sequencing of Therapy, Combinations, and Future Directions

Without question, the approvals of ipilimumab and vemurafenib marked a major advance in the treatment of locally advanced and metastatic melanoma. Within a little more than 1 year after the approval of these agents, compelling clinical data were presented for two investigational monoclonal antibodies against PD-1, nivolumab and MK-3475, which appear to be more effective and perhaps better tolerated than ipilimumab, and for the investigational combination of BRAF and MEK inhibitors, which appears to be better tolerated and more effective than treatment with a BRAF inhibitor alone. Moreover, high-dose IL-2 remains a viable option for selected patients because of its ability to induce durable remissions in a small subset, and select centers are able to offer trials of adoptive cellular therapy, which have shown substantial promise in phase II trials.

Assuming that anti-PD-1 antibodies and the combination of BRAF and MEK inhibitors become more widely available in the near future, clinicians and investigators will be faced with an array of active therapies, as well as with major questions regarding how to select and sequence therapies for individual patients. A major question for patients with tumor *BRAF* mutations will be which sequence of molecular targeted therapy and immunotherapy to administer in order to obtain the best survival outcome with the least toxicity. The relative rapidity of tumor response to molecular targeted therapy must be weighed against the current assumption that immunotherapy takes longer to produce response but may be more likely to produce longer and unmaintained remissions. However, the assumption of slower response to immune therapy may be challenged by agents such as anti-PD-1, or combinations that involve anti-PD-1, which may produce more rapid onset of tumor regression. The impact of a prior therapy on response and toxicity with a subsequent therapy—for example, a BRAF inhibitor followed by an immune therapy, or the sequencing of two immune therapies—remains mostly unknown. Current clinical experience indicates that resistance to one immune therapy does not preclude objective response to a subsequent immune therapy—for example, anti-PD-1 following ipilimumab, or ipilimumab following anti-PD-1.

Combination therapies offer the possibility of synergistic antitumor activity but may be complicated by increased or unexpected toxicities. Inhibitors of BRAF have been shown to increase tumor T-cell infiltration[74]—hence the rationale for combining these with immune therapies—but they could also enhance T-cell activation and toxicity through the paradoxical activation of C-RAF in normal cells. Combinations of certain immune checkpoint inhibitors, or checkpoint inhibitors combined with cytokines or immune costimulatory antibodies, may lead to more autoimmune adverse events. Nevertheless, combinations offer the greatest promise for further improvements in outcome, and each combination will be judged on its relative risk-benefit ratio and our ability to manage induced adverse events.

With more effective therapies and more combinations to test, it will be challenging to develop new agents for certain subsets of melanoma patients. Nevertheless, there are few effective therapies for patients with metastatic ocular melanoma, and there is no compelling effective molecular therapy for patients progressing after immune therapy who have tumor *N-RAS* mutations (approximately 15% of all melanoma patients) or tumors that do not have mutations in either *BRAF* or *N-RAS*. [75-78] Preliminary results from a phase II trial showed some activity for a MEK inhibitor in patients with *N-RAS* tumor mutations [79]; however, response durations were relatively short. Sequencing of the melanoma genome did not reveal additional common driver mutations amenable to rapid drug development. [80] Testing novel agents in the foregoing subsets of patients, including combinations of signaling pathway antagonists, new immune therapy agents, antibody-drug conjugates, and angiogenesis inhibitors, will be required to produce additional meaningful treatment advances in the near- to mid-term.

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