# Advances in Targeted Therapy for Melanoma

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**Abstract:** Metastatic melanoma remains an aggressive malignancy conferring a very poor prognosis, and standard chemotherapeutic and immunologic treatments have not demonstrated an overall survival benefit. No molecularly targeted therapy is approved for the treatment of advanced melanoma. Melanoma is a molecularly heterogeneous malignancy, and optimal treatment in a given patient is likely to depend on the presence of specific molecular abnormalities. Aberrations in components of signal transduction pathways have been identified that modulate melanoma proliferation and survival. Mutations that activate the mitogen activated protein kinase (MAPK) pathway via BRAF or NRAS are present in the majority of melanomas arising on skin intermittently exposed to the sun. Mutations that activate the KIT oncogene are more commonly present in melanomas arising from mucosal, acral, or chronic sun-damaged sites. Inhibitors of the MAPK pathway and of KIT are currently undergoing clinical investigation. In this article, we review advances in targeted strategies to treat different subgroups of patients with melanoma.

Keywords
Melanoma, BRAF, KIT, MAPK, MEK, GNAQ

## Background

Melanoma has traditionally been classified by site of origin and histologic features. The vast majority of melanomas originate as cutaneous lesions, with a minority developing from melanocytic structures in the eye or mucosal surfaces such as the sinuses, vagina, or anus. Approximately 30 years ago, classification systems were developed to group cutaneous melanomas into major clinical-histologic types, including superficial spreading, nodular, lentigo maligna, and acral melanoma.[1,2] Unfortunately, the usefulness of these divisions when treating patients with standard therapy for advanced melanoma remains minimal.

The American Joint Committee on Cancer staging system has recently been revised to allow for more accurate prognostication.[3] However, staging continues to depend upon both histologic features (depth of invasion and the presence or absence of ulceration or mitoses at the primary site) and the extent of spread (regional lymph nodes or distant spread). Molecular abnormalities within a given melanoma are currently not utilized for staging or the application of standard therapy.

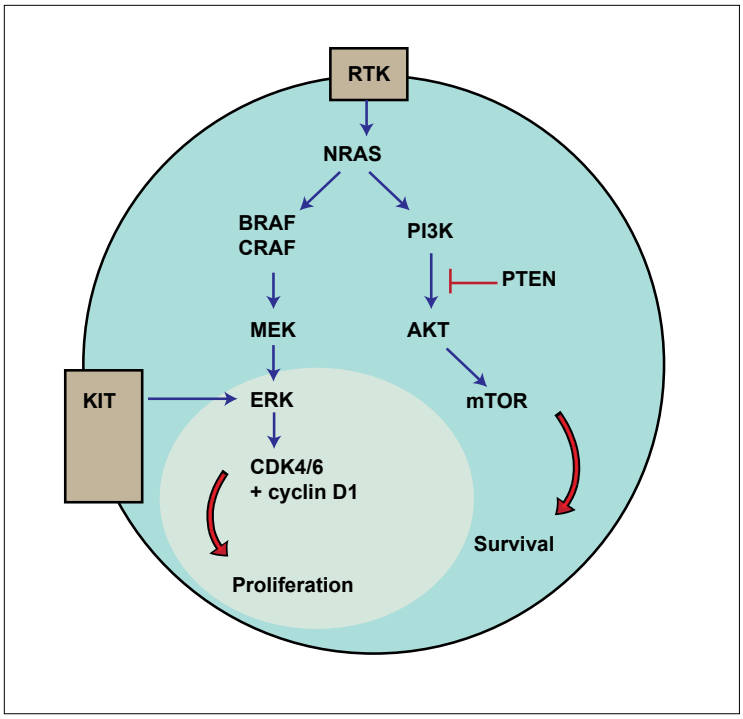


**Figure 1.** Dysregulation of signal transduction pathways in melanoma. The mitogen activated protein kinase and PI3K/akt pathways regulate proliferation and survival of melanocytes. Activating mutations in melanomas have been detected in BRAF, NRAS, and KIT. Other abnormalities detected include deletion of PTEN and increased expression of CDK4 and cyclin D1.

Throughout the previous decade, important advances have elucidated molecular and genetic abnormalities critical for melanoma proliferation and survival (Figure 1). We are increasingly realizing that melanoma is in fact a molecularly heterogeneous disease. Systematic genome-wide screening identified missense mutations in BRAF, a component of the mitogen activated protein kinase (MAPK) pathway in 66% of melanomas.[4] The most common BRAF mutation leads to constitutive activation of the kinase domain through substitution of glutamic acid for valine at position 600 (V600E). An additional 15–20% of melanomas contain activating mutations in NRAS, a component of the MAPK pathway upstream of BRAF. Since concurrent BRAF and NRAS mutations are rare, approximately 80% of melanomas have selected mutations that activate the MAPK pathway. A prospective cohort study of 251 patients with cutaneous melanoma found an association between the presence of a mutation in NRAS and increased proliferation at the primary site (as evidenced by deeper breslow thickness and increased mitoses) and shorter melanoma-specific survival when compared to melanomas containing a mutation in BRAF or wild-type for both BRAF and NRAS.[5]

The presence of activating mutations within the MAPK pathway was discovered in unselected melanoma populations. Subsequent genomic approaches that assessed the mutational status of BRAF and NRAS have identified 4 distinct sets of genetic alterations in melanoma.[6] Melanomas arising on chronically sun-exposed skin (histologically defined by the presence of solar elastosis), intermittently sun-exposed skin, acral surfaces, and mucosal surfaces differ in the frequency of BRAF and NRAS mutations. Eighty-one percent of melanomas arising on intermittently sun-exposed skin contain BRAF or NRAS mutations.

Mutations in BRAF and NRAS are less frequently detected in melanomas arising on chronically sun-damaged skin, acral surfaces, and mucosal surfaces. The presence of a sub-centromeric amplification in a region encompassing the KIT gene on chromosome 4q in acral melanoma led to speculation that activating abnormalities in KIT may play a role in the development of these melanomas.[7] Comparative genomic hybridization refined the boundary of the amplification and confirmed the presence of KIT within it. Strikingly, this amplification was essentially limited to melanomas lacking BRAF or NRAS mutations.[8] KIT is a tyrosine kinase whose activation regulates downstream signal transduction pathways including the MAPK pathway and the PI3K/akt pathway.[8,9] Sequencing of KIT identified mutations in 17% of melanomas on chronically sun-damaged skin, 11% of melanomas on acral surfaces, 21% of melanomas on mucosal surfaces, but less than 1% of melanomas on intermittently sun-exposed skin.[8]

The molecular heterogeneity of melanoma is associated with the location of the primary site (Table 1). The site of origin can help clinicians estimate the likelihood of specific genetic abnormalities. Recent advances in understanding ocular melanomas, for example, have

**Table 1.** The Frequency of Activating Mutations in Subgroups of Melanomas

| Primary Site | BRAF[6] | NRAS[6] | KIT* | GNAQ[52] |
|---|---|---|---|---|
| CSD† | 10% | 15% | 28% | |
| Non-CSD‡ | 60% | 20% | <1% | |
| Acral | 20% | 10% | 36% | |
| Mucosal | 10% | 5% | 39% | |
| Ocular | <1% | <1% | | 46% |

CSD=chronically sun damaged; GNAQ=guanine nucleotide binding protein, alpha q polypeptide.

*Mutations and/or copy number increases, Curtin JA et al. *J Clin Oncol.* 2006;24:4340-4346.

†Chronically sun-exposed skin as defined by the presence of solar elastosis.

‡Nonchronically sun-exposed skin.

immediate implications for therapeutic decisions. Strategies to inhibit aberrantly activated signal transduction are currently being investigated. In this article we review advances in targeted approaches to treating melanoma.

## Inhibition of the MAPK Pathway

The selection of activating NRAS and BRAF mutations in melanoma suggests that pharmacologic inhibition of the MAPK pathway may provide therapeutic benefit. This hypothesis is supported by a large body of preclinical data validating mutant BRAF or NRAS as potential therapeutic targets.[10-12] The efficacy of a targeted therapeutic strategy will depend on the selectivity and potency of the therapy to inhibit its target.

NRAS requires posttranslational modifications for activation. Inhibition of farnesylation prevents correct membrane localization and activation of RAS proteins.[13] A phase II study evaluating the single-agent activity of the farnesyltransferase inhibitor R11577 (Johnson & Johnson) demonstrated no responses in the 14 treated melanoma patients.[14] The poor activity seen with R11577 may be due to patient selection, as tumors were not analyzed for NRAS mutations.

Another approach to down regulate the MAPK pathway is through targeted inhibition of BRAF. Sorafenib (Nexavar, Bayer) was developed as a selective RAF inhibitor. Subsequent studies revealed that it is less selective and inhibits an array of other kinases including vascular endothelial growth factor receptors 1 and 2.[15] Although it inhibits the growth of V600E BRAF tumors in xenograph models, single-agent sorafenib demonstrated minimal activity in melanoma patients.[16] The addition of sorafenib to dacarbazine chemotherapy in a randomized phase II study of chemotherapy-naïve melanoma patients resulted in an encouraging improvement in median progression-free survival (PFS; 11.7 weeks to 21.1 weeks; $P$=.068) but not in overall survival.[17] A phase III study comparing carboplatin and paclitaxel chemotherapy alone to the combination with sorafenib as second-line treatment failed to demonstrate a significant progression free or overall survival benefit.[18] Similarly, a phase III study with 823 chemotherapy-naïve melanoma patients failed to demonstrate significant benefit in rate of response or survival with the addition of sorafenib to carboplatin and paclitaxel.[19] The reason for the limited activity seen with sorafenib is not entirely clear, but is likely due to suboptimal MAPK inhibition.[20] While sorafenib demonstrated efficacy leading to US Food and Drug Administration approval for the treatment of renal cell and hepatocellular carcinomas, the primary mechanism of benefit is likely through anti-angiogenic mechanisms and not through the inhibition of the MAPK pathway.[20]

Although sorafenib is the most extensively studied RAF inhibitor in melanoma, more selective and potent RAF inhibitors are in development. PLX4032 and PLX4720 (Plexxikon) are selective BRAF inhibitors. These agents have more than 10-fold greater selectivity for V600E BRAF compared to the wild-type protein (half maximal inhibitory concentration of 13 nM vs 160 nM for PLX4720).[21] PLX4032 is the most selective inhibitor of V600E mutant BRAF to enter clinical investigation.[22] These compounds were developed using a crystal structure–guided approach to synthesize compounds predicted to bind the kinase domain of V600E BRAF.[21] Selectivity was validated in preclinical models demonstrating inhibition of phospho-ERK expression in V600E, but not wild-type BRAF–expressing cell lines, and the inhibition of tumor growth selectively in V600E expressing tumors.[21]

A phase I dose escalation study with PLX4032 revealed promising activity in patients with V600E mutated but not wild-type BRAF–expressing melanoma. Nine of 16 patients with V600E BRAF–expressing melanoma and none of 5 patients with wild-type BRAF–expressing melanoma demonstrated a response. Although the data are thus far immature, the median PFS in subjects with mutated BRAF is approximately 6 months.[22] A phase II study of PLX4032 in patients with mutant V600E BRAF–expressing melanoma who previously received standard therapy has recently completed accrual. A phase III first-line study with randomization of PLX4032 to standard chemotherapy is currently accruing patients.

Similarly, a phase I/II study of patients with mutated BRAF–expressing melanoma who were treated at higher

doses of the selective BRAF inhibitor GSK2118436 (GlaxoSmithKline) demonstrated a 63% overall response rate. GSK2118436 may also have activity in the central nervous system, with responses seen in small, previously untreated brain metastases in several patients.[23] Other RAF inhibitors, such as RAF-265 developed by Novartis and XL281 developed by Exelixis ,are in phase I development.

An alternative strategy to inhibit the MAPK pathway involves inhibition of targets downstream of RAF. AZD6244 (Array Biopharma/AstraZeneca) is a potent and selective inhibitor of MEK1/2. Inhibition of MEK1/2 is an interesting possibility given its only known target is ERK, a component of the MAPK pathway downstream from RAF. In preclinical melanoma models, AZD6244 demonstrated largely cytostatic effects with tumor regression seen only in conjunction with chemotherapy.[24] Preclinical activity has been observed in both mutated BRAF and mutated NRAS melanoma models.[25] Phase I data demonstrated anti-tumor activity in melanoma.[26] However, a phase II study with the primary endpoint of PFS in patients treated with AZD6244 or temozolomide demonstrated no significant difference between the 2 arms or in the mutated BRAF subgroup. Only 12% of patients with mutated BRAF obtained a partial response.[27] The modest activity seen may be due to suboptimal pharmacokinetics of AZD6244. It has been suggested that the limited activity also could be the result of compensatory mechanisms in the setting of an agent that is largely cytostatic in preclinical models.[24] Within V600E BRAF–mutated melanomas, genetic heterogeneity exists with alterations of other components of the MAPK pathway or of additional signaling pathways. Examples include PTEN, cdk4, p16, akt, and MITF.[6,28]

Other MEK inhibitors are in development, with results from phase I studies recently presented. Three of 8 melanoma patients treated with the oral MEK inhibitor AS703026 (Merck Serono) demonstrated partial responses, but the relationship between response and BRAF or NRAS mutation status remains unknown.[29] Treatment of 20 patients with melanoma expressing a BRAF mutation with the MEK1/2 inhibitor GSK1120212 (GlaxoSmithKline) resulted in a 40% response rate with 2 complete and 6 partial responses and only 4 patients demonstrating early progression. Efficacy was lower in patients with melanoma expressing wild-type BRAF, as only 2 of 22 patients demonstrated a partial response.[30] A phase I study of the MEK1 inhibitor E6201 (Eisai) determined the maximal tolerated dose, and accrual to an expansion cohort of patients with melanoma expressing a BRAF mutation is under way.[31]

An important area of active investigation involves the identification of resistant mechanisms to effective BRAF inhibition in V600E mutant tumors. An in vitro MEK1 random mutagenesis screen identified mutations that conferred resistance to MEK and BRAF inhibition.[32] One such mutation in MEK1 was detected in a resistant metastatic focus in a melanoma patient treated with AZD6244. Interestingly the exposure of V600E mutated BRAF-containing cell lines to direct inhibitors of both BRAF and MEK prevented the emergence of resistant clones.[32] In preclinical models, amplification of cyclin D1 (detected in 17% of mutant BRAF samples) also increased resistance to BRAF inhibition.[33]

As RAF proteins are upstream of MEK, inhibition of BRAF could potentially disrupt signaling to other downstream substrates, leading to a different functional outcome than that seen with the inhibition of MEK. However, microarray analyses of V600E mutant BRAF–expressing melanoma cell lines exposed to either a BRAF or MEK inhibitor found that both inhibitors altered the expression of a common set of genes.[34]

Although RAF inhibition failed to demonstrate MEK independent effects on transcriptional output, the clinical efficacy of inhibiting the MAPK pathway is likely to be dependent upon the presence or absence of a mutation in BRAF or NRAS and on the component of the pathway being inhibited. Exposure of a panel of cell lines to the RAF inhibitor PLX4032 inhibited proliferation only in melanomas containing a V600E mutation in BRAF. Cell lines containing a mutation in NRAS or wild-type BRAF and NRAS were resistant to RAF inhibition. Similarly, exposure of these cell lines to a MEK inhibitor arrested the proliferation of melanomas expressing a V600E mutation in BRAF but not wild-type BRAF. Melanomas with a mutation in NRAS displayed variable sensitivity to MEK inhibition, with a subset of cell lines that were insensitive to RAF inhibition demonstrating growth arrest upon inhibition of MEK.[34]

During evaluation of downstream effects on the MAPK pathway in melanoma cell lines that contain a V600E BRAF mutation, it was noted that inhibition of RAF with PLX4032 inhibited the activity of MEK and ERK. Unexpectedly, exposure of melanoma cell lines that express wild-type BRAF to PLX4032 induced MEK and ERK signaling. Mechanistically, this downstream activation is dependent upon the binding of one member of a CRAF-CRAF or CRAF-BRAF dimer to PLX4032, with activation of the drug-free member.[35] BRAF inhibition as a single-agent treatment strategy should be studied with caution in patients with melanoma lacking a BRAF mutation, given the potential to paradoxically activate downstream components of the MAPK pathway.

Initial studies with melanomas resistant to RAF inhibition also have implicated increased CRAF expression as a mechanism of resistance.[36] However, resistance mechanisms are likely to be complex and result from

dysregulation of multiple signaling pathways and alternative components within a given pathway. Elucidation of these compensatory mechanisms is critical to develop more effective treatment approaches. The inhibition of multiple targets may likely be necessary for optimal therapeutic benefit.[37]

Melanomas containing NRAS mutations or BRAF mutations with concurrent loss of PTEN function could potentially be sensitive to dual MAPK and PI3K/akt pathway inhibition. Phospho-akt expression increases with melanoma progression and is inversely correlated with survival.[38] Dual inhibition of MAPK and PI3K/akt pathways induced cell death and inhibited cellular proliferation in preclinical melanoma models.[37,39] Potent inhibitors of akt and PI3K are now in clinical phase I development.

## KIT

Activating mutations in KIT were initially reported in 21% of mucosal melanomas, 11% of acral melanomas, and 16.7% of melanomas arising on chronically sun-damaged skin.[8] Additional cases demonstrated increased copy number or amplification of KIT, with or without the presence of mutations. Such changes rarely develop in melanomas arising on cutaneous surfaces lacking solar elastosis. In mucosal melanoma cell cultures containing mutated KIT, treatment with the KIT inhibitor imatinib (Gleevec, Novartis) caused inhibition of both the MAPK and PI3K/akt pathways.[40]

Three phase II studies evaluating the efficacy of KIT inhibition with imatinib in unselected populations of melanoma patients demonstrated a lack of clinical efficacy.[41-43] However, KIT inhibition was subsequently validated as a therapeutic target in melanomas that selected for activating KIT mutations. Two case reports of melanoma patients with KIT mutations demonstrated dramatic activity following treatment with imatinib.[40,44] Preliminary results from a phase II study of imatinib in patients with KIT-mutated or KIT-amplified melanoma demonstrated responses in 4 of 12 evaluable patients.[45] Two patients developed a complete response, 2 a partial response, 6 stable disease, and 2 progressive disease. Two of the responses were durable, lasting more than 37 weeks.

Proper patient selection is important when considering KIT-directed therapy. Increased expression of KIT by immunohistochemistry (IHC) does not correlate with the KIT mutational status of the tumor. A phase II study of imatinib in patients with melanoma that expressed KIT by IHC demonstrated activity in only 1 of 21 patients.[43] Similarly, melanomas containing a KIT mutation do not always express KIT by IHC.[46,47]

It remains unclear whether there is benefit with KIT inhibitors in patients whose tumors express amplified wild-type KIT. Furthermore, the duration of response, progression free or overall survival benefits, and mechanisms of resistance need to be determined in responding patients and then in those with de novo resistance. From experiences with KIT inhibition in gastrointestinal stromal tumors (GIST), one would propose that a possible mechanism of resistance would be the acquisition of additional mutations in different KIT exons. The optimal KIT inhibitor for a given patient may depend upon on the specific KIT mutation. For example, a melanoma cell line harboring an L576P mutation in KIT demonstrated decreased cell viability upon exposure to dasatanib (Sprycel, Bristol-Myers Squibb) but not imatinib or nilotinib (Tasigna, Novartis).[48] Correlation of specific KIT mutations and prior treatments to responsiveness to a particular tyrosine kinase inhibitor, however, still requires additional clinical investigation. Phase II studies are under way with KIT inhibitors, including sunitinib (Sutent, Pfizer), nilotinib, and dasatinib. Correlation of efficacy with the specific KIT mutations will be important to guide agent selection.

## Ocular Melanoma

Melanomas arising from melanocytic structures in the eye respond poorly to chemotherapy and immunotherapy. While MAPK pathway activation has been demonstrated, these melanomas invariably lack BRAF and NRAS mutations.[49] Although specific cytogenetic alterations have been associated with these melanomas, until recently, abnormalities in specific oncogenes and tumor suppressor genes have not been definitively implicated in tumor progression.[50] A genetic screen in mice identified mutations in GNAQ and GNA11 that cause diffuse skin hyperpigmentation due to an increase in the number of intradermal melanocytes.[51] GNAQ and GNA11 are members of a family of G-protein alpha subunits necessary for G-protein–coupled receptor signaling to downstream effectors. Systematic sequencing of the coding regions of GNAQ in 236 melanomas demonstrated GNAQ mutations in 46% of ocular melanomas.[52] The mutations develop at codon 209 within the Ras-like domain resulting in constitutive activation. An additional 30% of ocular melanomas have GNA11 mutations (BC Bastian, MD, PhD, University of California, San Francisco, personal communication). These mutations lead to constitutive activation of the MAPK pathway.[52,53] Inhibition of GNAQ expression using siRNA against GNAQ in ocular melanoma cell lines results in inhibition of the MAPK pathway and G1 cell cycle growth arrest.[52] As a result, this provides a rationale to investigate the efficacy of MAPK pathway inhibition in ocular melanomas. Clinical trials designed to evaluate the efficacy of MEK inhibition in this population are planned.

***Combination of Targeted Therapy With Chemotherapy***

It remains unclear what specific interactions exist between MAPK pathway activity and the cytotoxic effects of chemotherapy. MEK can upregulate various DNA repair genes potentially dampening the cytotoxic effect of chemotherapy.[54] Taxanes are known to activate the MAPK pathway. MEK activity is required to complete mitosis. Combined MEK inhibition and taxane treatment results in additive growth arrest and cell death in preclinical models. In melanoma cell lines, the MEK inhibitor AZD6244 enhances the apoptotic effects of docetaxel.[24] Phase II data suggest that the nonselective BRAF and CRAF inhibitor sorafenib may enhance the activity of dacarbazine.[17] Comparative studies of the combination of chemotherapy and MEK or BRAF inhibition versus chemotherapy alone versus MEK or BRAF inhibition alone would permit clinical validation of chemosensitizing effects through MEK inhibition. In a small nonrandomized study, treatment with the combination of AZD6244 and chemotherapy resulted in a 56% response rate in patients with tumors expressing a mutation in BRAF but no response in patients with tumors containing a mutation in NRAS or wild-type BRAF and NRAS.[55] The rate of response in the subset of patients with a mutation in BRAF was greater than expected for either agent alone, suggesting augmentation of the efficacy of chemotherapy through MEK inhibition. To further test this hypothesis, a phase II randomized study assessing the efficacy of AZD6244 in combination with dacarbazine in comparison to dacarbazine alone in V600E BRAF–expressing melanoma is under way.

The cytotoxic effects of chemotherapeutic agents are mediated by modulation of the activity of apoptotic pathways. Resistance to chemotherapy in melanoma has been linked to overexpression of the antiapoptotic protein Bcl-2.[56] Oblimersen sodium is an antisense oligonucleotide that downregulates Bcl-2 protein expression and sensitizes human cancer cells to chemotherapy-induced apoptosis in xenograph models.[57] A phase III study (GM301) in chemotherapy-naïve patients demonstrated a significant PFS benefit with the addition of oblimersen sodium to dacarbazine.[58] Subset analysis identified a significant interaction between baseline serum LDH and treatment. Oblimersen sodium significantly improved overall survival in the subset of patients with normal baseline serum LDH but not in the study population as a whole. The survival benefit appreciated on subset analysis led to the development of AGENDA (Trial of Dacarbazine With or Without Genasense in Advanced Melanoma), a second phase III study assessing the efficacy of oblimersen sodium in combination with dacarbazine specifically in patients with low baseline serum LDH. This study has completed accrual, with ongoing follow-up for overall survival. Pooled analysis of the GM301 and AGENDA studies demonstrated modest improvement in overall response rate and PFS with the addition of oblimersen to dacarbazine, but overall survival data remain immature.[59]

The addition of antiangiogenic agents to chemotherapy has resulted in improved efficacy and tumor control in a variety of malignancies.[60,61] Vascular endothelial growth factor (VEGF) is an endothelial mitogen that mediates angiogenesis. Downregulation of VEGF activity in human melanoma xenograph models inhibited tumor cell growth.[62] Bevacizumab (Avastin, Genentech) is a monoclonal antibody that blocks angiogenesis by binding to VEGF, preventing coupling to the VEGF receptor.[63] The addition of bevacizumab to carboplatin and paclitaxel in a randomized phase II study of previously untreated advanced melanoma patients demonstrated an improvement in the primary endpoint of median PFS (5.6 vs 4.2 months) that did not reach statistical significance. A trend towards improved overall survival with the addition of bevacizumab (12.3 vs 9.2 months) was appreciated, but also did not reach statistical significance. Though these trends are encouraging, further investigation is necessary to optimize targeted anti-angiogenic therapeutic approaches and determine efficacy in subpopulations of melanoma patients.

## Future Directions

Mutational activation of members of the MAPK pathway and of KIT in select melanomas provides a strong rationale for targeted investigational strategies (Table 2). Therapeutic approaches can be tailored to the mutational status of BRAF, NRAS, and KIT in a given patient's melanoma. Individuals whose melanoma developed on chronically sun-exposed, acral, or mucosal surfaces should have their tumor evaluated for activating KIT mutations with the anticipation of treatment with a KIT inhibitor.

Furthermore, patients whose melanomas developed on intermittently sun-exposed skin or on chronically sun-damaged skin but lack a KIT mutation should have their tumors evaluated for activating mutations of the MAPK pathway. Patients whose tumor harbors BRAF mutations should have the opportunity to participate in one of the many clinical trials of BRAF inhibitors under development. The specific BRAF mutation has significant implications. Preclinical data suggest that melanomas with non V600E BRAF mutations are reliant upon CRAF for survival.[64] Melanomas with NRAS mutations switch their downstream signaling from BRAF to CRAF as a result of disrupted cyclic AMP signaling.[65] One would predict these melanomas to be less sensitive to BRAF inhibition relative to CRAF inhibition. Selective means to inhibit CRAF could reveal greater promise in NRAS mutated or non-V600E BRAF–mutated melanomas.

**Table 2.** Examples of Targeted Strategies Under Investigation to Treat Melanoma

| Target/mechanism | Agent | Company | Trial Status in Melanoma |
|---|---|---|---|
| N-RAS/farnesyltransferase inhibitors | R11577 | Johnson and Johnson | Phase II |
| Nonselective BRAF/CRAF tyrosine kinase inhibitors | Sorafenib | Bayer/Onyx | Phase III with carboplatin and paclitaxel |
| BRAF inhibitors | PLX4032 | Plexxikon/Roche | Phase III randomized versus dacarbazine |
| | RAF265 | Novartis | Phase I |
| | GSK2118436 | GlaxoSmithKline | Phase I |
| MEK/tyrosine kinase inhibitors | AZD6244 | AstraZeneca | Phase II |
| | GSK1120212 | GlaxoSmithKline | Phase II in patients with mutant BRAF previously treated with BRAF inhibitor |
| | PD0325901 | Pfizer | Phase I |
| | AS703026 | Merck Serono | Phase I |
| | E6201 | Eisai Co. | Phase I |
| KIT/tyrosine kinase inhibitors | Imatinib | Novartis | Phase II in patients with amplified or mutant KIT |
| | Sunitinib | Pfizer | Phase II in patients with acral or mucosal melanoma |
| | Nilotinib | Novartis | Phase II in patients previously treated with KIT inhibitor. Phase III randomized versus dacarbazine. |
| | Dasatinib | Bristol-Myers Squibb | Phase II in patients with mutant KIT |

Despite the tremendous recent advances in our understanding of melanoma signaling biology, many unanswered questions remain. Specifically, whether a survival benefit exists for such targeted therapy, duration of treatment responses and mechanisms of resistance need to be determined. Melanoma progression likely generates redundancy between numerous signal transduction pathways. As a result, it may be necessary to inhibit multiple signaling pathways to generate durable and improved clinical benefits.

One theorized treatment strategy to investigate is combined inhibition of the PI3K/akt and the MAPK pathways. Approximately 30% of melanoma cell lines contain deletion in PTEN function, and 50% demonstrate constitutive activity of akt3.[66] Combined inhibition of akt3 and mutant BRAF using siRNA technology retards melanoma xenograft growth in nude mice.[67] Inhibitors of akt and PI3K are in clinical development and could potentially be combined with BRAF or MEK inhibitors to test such a hypothesis.

An alternative strategy to obtain dual pathway inhibition of MAPK and akt is through treatment with heat shock protein 90 (hsp90) inhibitors. Hsp90 is a chaperone protein that binds to and stabilizes the conformation of client proteins. These clients include wild-type and mutant BRAF, CRAF, akt, cdk4, and KIT.[68] Potent hsp90 inhibitors currently in development have the potential to inhibit both the PI3K/akt and MAPK pathway and provide another means of clinical investigation.[69]

The discovery of activating mutations in large subsets of melanomas has led to the testing of rational targeted therapeutic approaches. Treatment planning for advanced disease is shifting from a focus on histologic features to molecular abnormalities. Melanomas are genetically heterogeneous with redundancy among multiple signal transduction pathways. Effective and durable targeted treatment approaches will likely require inhibition of multiple pathways or combinations with cytotoxic agents. Therapeutic design may one day be tailored to the specific genetic and molecular profile of a given patient's melanoma. As more potent and selective agents are developed, there will be a greater ability to test these hypotheses with hopeful translation into improved patient outcomes.

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