Chemotherapy Resistance Abrogation in Metastatic Melanoma

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Keywords
Drug resistance, melanoma, O^6-methylguanine-methyltransferase, mismatch repair, drug development, PARP inhibitor

Abstract: Melanoma is rapidly increasing in incidence throughout the world. Based on American Cancer Society estimates, there will have been approximately 68,720 new cases of invasive melanoma diagnosed in 2009 in the United States. The increase in melanoma incidence has not been paralleled by the development of new therapeutic agents with a significant impact on survival. The promise of targeted therapy has not yet been brought to bear, making chemotherapy with alkylating agents the mainstay of therapy of metastatic melanoma despite the dismally low response rates. The resistance of tumors to these agents is in part due to DNA repair mechanisms that allow cells to survive alkylation damage. Several novel agents targeting the abrogation of DNA repair pathways alone and in combination with cytotoxic agents have been developed with varying measures of success. This review summarizes the current knowledge of the dysregulation of DNA repair pathways as mechanisms of resistance to chemotherapy in melanoma and their potential as targets for novel developmental therapeutics.

Introduction

The incidence of melanoma continues to rise at an alarming pace. Substantial proportions of patients develop metastatic disease and eventually succumb to it within a year. Based on American Cancer Society estimates, there will have been approximately 68,720 new cases of invasive melanoma diagnosed in 2009 in the United States, which resulted in approximately 8,650 deaths in 2009.[1] The survival rate at 5 years for patients with metastatic disease is less than 10% with a median survival of 6–9 months.[2] Unfortunately, the increase in incidence has not been paralleled by the development of new therapeutic agents with a significant impact on survival.[3,4] The promise of targeted therapy has not yet been brought to bear, making chemotherapy with alkylating agents the mainstay of therapy of metastatic melanoma despite the dismally low response rate of 10% at best.[5]

Dacarbazine is the only approved chemotherapeutic agent for the treatment of metastatic melanoma.[5] Among the 8 randomized trials in which dacarbazine was used as a comparator arm since

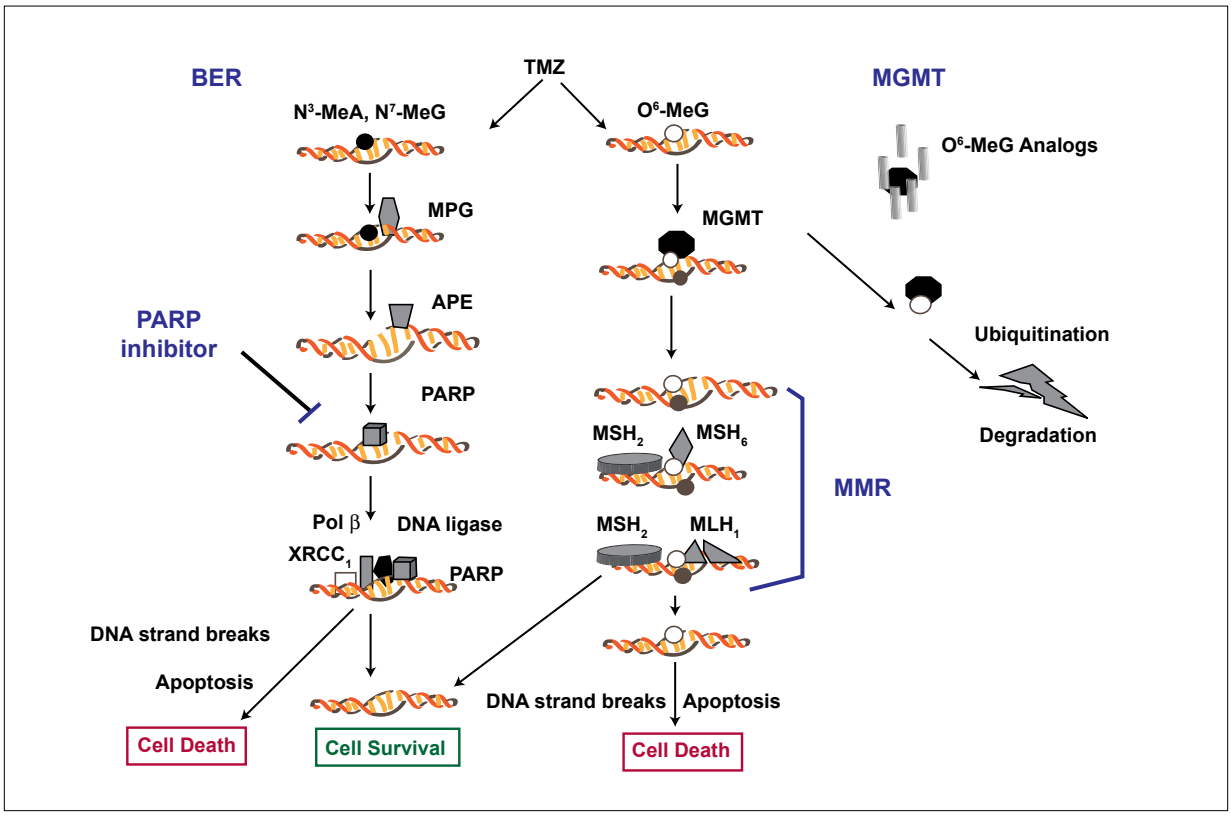

Figure 1. Three DNA repair pathways are involved in reversing the DNA damage induced by temozolomide (TMZ). TMZ methylates DNA at the O^6- position of guanine (resulting in O6-methylguanine (O6-MeG), N7-methylguanine (N7-MeG), and N3-methyladenine (N3-MeA). N7-MeG and N3-MeA DNA adducts (>70% of total DNA adducts induced by TMZ) are rapidly and efficiently repaired by the base excision repair (BER) pathway, and normally contribute little to TMZ-induced cell death. Inhibition of poly-(ADP)-ribose polymerase (PARP) of the BER pathway and accumulation of double-strand breaks that eventually culminate in cell death. MGMT (O^6-meG DNA methyltransferase) removes the O^6-alkylguanine DNA adduct but is inactivated and degraded in the process. A similar reaction occurs when MGMT transfers and accepts an alkyl-group from O^6-MeG analogs. Mismatch repair (MMR) recognizes the mis-pair formed during replication of an un-repaired O^6-MeG lesion. MMR attempts to repair this mismatch but fails, resulting in futile cycles of repair that then initiate cell death. Cancer cells down-regulate the MMR system through epigenetic silencing to evade alkylator-induced cell death.

1992, more than 1,000 patients have been treated with dacarbazine and show an overall response rate of 13.4%.[6] The oral agent temozolomide is rapidly converted to the same methylating active moiety as dacarbazine and, with its favorable side-effect profile, is widely used for the treatment of metastatic melanoma. Randomized trials have confirmed that temozolomide is at least equivalent to dacarbazine in terms of efficacy, although temozolomide had a better side effect profile and has shown modest improvements in health-related quality of life in a phase III trial conducted by Middleton and colleagues.[5,7]

Other cytotoxic drugs with activity in melanoma include the nitrosoureas,[8] platinum analogs,[9,10] vinca alkaloids,[11] and taxanes.[12] Recently, the combination of carboplatinum with paclitaxel has shown activity in melanoma in phase II and III trials, though the response rates were not higher than 20%.[13,14] This finding has led to the wide adoption of this regimen as one of the standard therapies for patients with melanoma. It is noteworthy that the same regimen can achieve response rates as high as 60% or even 80% in epithelial malignancies such as lung, breast, and ovarian cancers.

This decreased benefit highlights melanoma as a prototype of chemotherapy-resistant tumors. The mechanisms underlying such resistance appear to be closely tied to dysregulation of DNA repair pathways in cancer cells. DNA repair pathways (Figure 1) are present in normal cells to maintain genome integrity.

Multiple pathways are activated in response to the genotoxic damage induced by chemotherapy, starting with recognition of DNA damage and culminating in programmed cell death.[15] Failure at any point along this cascade of events can be translated into resistance.

This review will recapitulate the current knowledge of the dysregulation of DNA repair pathways as mechanisms of resistance to chemotherapy in melanoma and their abrogation as a target for novel developmental therapeutics.

O^6-Methylguanine-Methyltransferase

O^6-Methylguanine-methyltransferase (MGMT) is one of the most studied mechanisms of resistance to alkylating agents. It is a ubiquitously expressed and highly conserved DNA repair protein that is vital in the maintenance of DNA integrity. Unlike other DNA repair mechanisms, MGMT does not activate a pathway but is a single-protein pathway that recognizes and repairs DNA damage through its specificity for O^6- substituted purines.[16] MGMT recognizes the O^6-guanine base lesion induced by alkylating agents and transfers the methyl group to a cysteine residue in its active site. The guanine base is therefore repaired and can sustain regular replication and transcription, while the MGMT molecule is inactivated, ubiquitinated, and degraded.[15] Any additional DNA repair requires de novo synthesis of the protein.[17] This characteristic makes MGMT an ideal target for inhibition and in fact represented the first DNA repair-targeted resistance abrogation strategy.[16]

O^6-Methylguanine (O^6-MeG) analogs were developed with the goal of depleting MGMT by presenting it with decoy base lesions that are themselves devoid of toxicity. This strategy was successful at depleting MGMT in preclinical models. O^6-benzyl guanine (O^6-BeG) was the first agent to reach clinical investigation, and was used in combination with carmustine (BCNU), BCNU in wafer form, and temozolomide for treatment of different solid tumors such as lymphomas, gliomas, melanomas, sarcomas, and colon cancer.[18-26] Two phase I trials conducted at the University of Chicago and Case Western Reserve University[18,19] evaluated toxicity in patients with advanced solid tumors or lymphoma. Patients received O^6-BeG intravenously followed 1 hour later by carmustine. The University of Chicago trial determined that the maximum tolerated dose (MTD) of carmustine when combined with 120 mg/m^2 O^6-BeG was approximately 3-fold lower (40 mg/m^2) than the standard clinical dose.[19] Increased hematologic toxicity was the most significant adverse event associated with the addition of O^6-BeG to carmustine. In both studies, MGMT activity was successfully inhibited in peripheral blood mononuclear cells and even in tumor tissues in the Case Western Reserve University study.[18,19]

Increased myelosuppression continued to plague the development of this agent even in phase II trials; several patients with melanoma treated on a phase II trial of O^6-BeG and BCNU at 40 mg/m^2 required additional dose reduction on the basis of hematologic toxicity.[25] Despite the pharmacodynamic effects of MGMT depletion in peripheral blood mononuclear cells (PBMCs) manifested by significant myelosuppression, the clinical outcome was not improved. This experience was reproduced in several phase II trials in patients with other conditions, such as soft tissue sarcoma, multiple myeloma, and glioblastoma multiforme (GBM), where the increased toxicity was not associated with a comparable increase in efficacy.[26-28] This outcome necessitated re-examination of this strategy, particularly as it was determined that: a) MGMT levels rapidly recovered within 24–48 hours and b) that the total dose of alkylating agents delivered was curtailed by the exquisite sensitivity of bone marrow cells and subsequent myelosuppression.

O^6-(4-Bromothenyl)-guanine (lomeguatrib) is a next-generation O^6-MeG analog that is orally bioavailable and therefore offers the opportunity to surpass the main obstacle, as oral administration allows for versatility of scheduling and therefore a more prolonged inhibition of MGMT. In a phase I trial in patients with melanoma conducted at our institution, lomeguatrib was administered with dacarbazine daily for 5 days and escalated to twice daily for 10 days. Although this regimen was successful in prolonging MGMT inhibition, the MTD of dacarbazine was only 400 mg/m^2, less than 50% of the standard clinical dose (800–1,000 mg/m^2).[29] In a recapitulation of the O^6-BeG experience, no improvement in the efficacy of dacarbazine was observed, although a formal phase II trial is yet to be conducted. In a similar phase I trial of temozolomide (75 mg/m^2) and lomeguatrib (40 mg) conducted by Middleton's group in the United Kingdom, similar hematologic toxicity and limited clinical efficacy were seen, suggesting no advantage for this regimen over conventional temozolomide administration in the treatment of melanoma.[30]

A low-dose, extended-schedule administration of an alkylating agent was explored with oral temozolomide. Extended-schedule temozolomide offered a more sustainable (although less profound) MGMT inhibition, while the total delivered dose of the alkylating agent exceeded the standard clinical doses usually administered over a 5-day regimen. In a study by Tolcher and co-authors, temozolomide was given on a day 7, 14, and 21 schedule, with mean MGMT activity decreasing by 72%, 63%, and 73% after 7, 14, and 21 days of treatment, respectively.[31] Furthermore, temozolomide dose intensity on

the dosing schedule of 7 days every 2 weeks was 2.1- to 2.8-fold higher than that achieved with its approved dose-schedule (2,100 mg/m^2 vs 750–1,000 mg/m^2 every 4 weeks).[31] This strategy was the basis for a large European Organization for Research and Treatment of Cancer randomized phase III trial in metastatic melanoma in which 859 patients were randomized to receive temozolomide 150 mg/m^2/day orally on days 1–7 repeated every 14 days ("week on–week off") or dacarbazine 1,000 mg/m^2 intravenous (IV) every 21 days. The preliminary results, reported at the European Society for Medical Oncology, revealed a minor increase in response rates (10% vs 14%), although the extended-schedule temozolomide did not impart any survival benefit.[32]

The contribution of MGMT to melanoma resistance to methylating agents seems to be more dependent on downstream pathways that are capable of recognizing the persistent O^6-guanine base damage and initiating apoptosis. Promoter methylation of MGMT (leading to decreased MGMT expression) is a recognized predictor of improved response to temozolomide-based chemotherapy in patients with GBM.[33] The role of MGMT (or its promoter) as a predictive marker of response to alkylator-based chemotherapy in melanoma is much less defined and may in fact be more valuable for the prediction of toxicity.[34]

Mismatch Repair Pathway

The mismatch repair (MMR) pathway corrects base substitution mismatches and small loops generated during DNA replication. Inactivation of MMR leads to the accumulation of mutations, particularly in repetitive sequences such as microsatellites. This is seen as microsatellite instability (MSI).[35] Inherited mutations in MMR genes underlie the predisposition to colorectal tumors in the familial syndrome hereditary nonpolyposis colon cancer (HNPCC). HNPCC tumors display MSI. MSI is also quite common in some apparently sporadic tumors, including sporadic colorectal tumors (15% incidence) and, to varying extents, tumors of several other organs.

The lack of clinical efficacy observed with the single pathway inhibition of MGMT could be, in part, due to the dependence of this pathway on a functional MMR system for cytotoxicity to occur.[36] MGMT and MMR have contrasting effects on DNA O^6-MeG. The former provides an efficient mechanism of repair, whereas MMR does not remove the methylated base but transforms the latter into a lethal lesion and activates the apoptotic pathways. If MMR is deficient, the O^6-MeG lesion can persist without leading to apoptosis, and consequently the cell will survive. MMR deficiency is therefore an efficient mechanism of resistance. MMR deficiency occurs primarily through epigenetic silencing of the key MMR genes by promoter methylation. It was shown to be a reversible process through treatment with epigenetic agents, such as decitabine. Decitabine is a DNA-methyltransferase-1 (DNMT-1) inhibitor that is approved for the treatment of myelodysplastic syndromes, and if used at low doses, leads to significant DNA hypomethylation.[37]

Decitabine has been reported to induce hypomethylation in tumor xenografts, which are associated with increased sensitivity to carboplatin.[38] A recently reported phase I clinical trial of decitabine in combination with carboplatin determined the phase II recommended dose to be decitabine IV at 90 mg/m^2 (day 1) followed by carboplatin IV at an area under curve 6 (day 8) every 28 days. Decitabine produced a reduction in DNA methylation equivalent to or greater than that observed in the xenograft model.[39]

The loss of MMR is not commonly described in melanoma; MMR gene mutations and MSI are also infrequent.[40] The loss of MMR expression with increasing Clark levels has been reported in melanoma specimens.[41] This finding has been proposed as a mechanism for acquired resistance in some patients. Studies on melanoma cell lines that demonstrate acquired chemoresistance show loss of MMR proteins in 30–70% of cases.[42]

Treatment with decitabine in melanoma cells has also been reported to lead to re-expression of products of epigenetically silenced genes such as the MMR protein human mutL homolog1 (hMLH1), and, therefore, leads to a proficient MMR system sensitizing melanoma cells to the cytotoxic effects of chemotherapy.[38,43]

At the University of Pittsburgh, we are conducting a phase I/II clinical trial with decitabine in combination with extended-schedule temozolomide in patients with metastatic melanoma. Extended-schedule temozolomide is expected to deplete MGMT, whereas treatment with decitabine will lead to the re-expression of MMR proteins. It is, to our knowledge, the first attempted dual DNA repair inhibition approach in the clinic.

Base Excision Repair Pathway

The 2 pathways discussed above (MGMT and MMR) are mostly involved in the repair of the O^6-MeG base lesion. The O^6-MeG lesion is credited with most of the methylating agent cytotoxicity, although it represents less than 10% of all base lesions.[7,44] Over 80% of the remaining lesions are at the N^7 position of the guanine and are efficiently and rapidly repaired by the base excision repair (BER) pathway.[4] Inhibition of BER might, therefore, enhance the clinical efficacy of methylating agents and has been explored primarily through poly(ADP-ribose) polymerase (PARP) inhibition.

PARP

The PARP family of proteins is characterized by the enzymatic property of poly(ADP-ribosylation). This reaction uses nicotinamide adenine dinucleotide (NAD)+ as a substrate and catalyses the addition of long, branching chains of poly(ADP-ribose) polymers to target proteins. Such poly(ADP-ribosylation) modulates the catalytic activity and protein-protein interactions of these targets and thus influences a wide range of cellular processes. PARP-1 is responsible for at least 80% of total cellular PARP activity, and together with its nearest relative PARP-2, constitutes the DNA damage response arm of the PARP family.

Chemical inhibitors of PARP have been in the laboratory for decades, and the earliest compounds were analogs of the nicotinamide component of NAD+. These inhibitors competed with NAD+, which is the substrate for PARP's catalytic function, and prevented subsequent synthesis of poly(ADP-ribose). Since then, an impressive array of more potent inhibitors has been developed, all of which act in primarily the same way. PARP functions to enhance repair and suppress the potential formation of double strand breaks in the presence of damaged DNA.[45] The reaction catalyzed by PARP consumes NAD+. Since PARP is abundant and rapidly activated by DNA damage, high doses of DNA-damaging agents have been shown to reduce cellular NAD+ levels by 80% within 5–15 minutes, and the resulting lack of ATP renders cells unable to undergo the process of apoptosis. As a result, cell death occurs due to necrosis, leading to a cascade of inflammatory events. Pre-treatment with a PARP inhibitor prevents the depletion of NAD+ and enables cells to undergo apoptosis. Therefore, despite the fact that DNA repair is impaired, PARP inhibition may reduce the overall damage to tissues or organs exposed to massive inflammatory insults.[46]

In a proof-of-principle seminal paper published in *Nature*, Farmer and colleagues reported the exquisite sensitivity of BRCA deficient cells to PARP inhibition.[47] The explanation lies in the concept of "synthetic lethality," which occurs when there is a potent and lethal synergy between 2 nonlethal events. In this case, a highly specific PARP inhibitor induces a DNA lesion as well as a genetic loss of function for the DNA repair pathway required to repair the lesion, which is restricted only to the tumor. The mechanism by which this process occurs is postulated to be the abundant formation of double strand breaks in the absence of PARP activity and the presence of a dysfunctional homologous recombination pathway required for double strand breaks repair.[48]

Several different PARP inhibitors have been shown to increase the cytotoxic effects of alkylating agents, such as temozolomide, and topoisomerase I poisons, such as irinotecan.[49] There is less evidence to support the effect of PARP inhibition on enhanced tumor responses when treated with DNA cross-linking agents, such as cisplatin. The mechanisms underlying these effects have not been fully understood and are likely to vary according to the properties of the cytotoxic agent. Recently, Donawho and associates have reported that the PARP inhibitor ABT-888 (Abbott Laboratories) in combination with platinum drugs or cyclophosphamide, but not alone, causes regression of BRCA1-deficient MX-1 xenografts.[50] In a recent phase I trial in which 60 patients were treated with AZD2281, a PARP1 inhibitor, objective antitumor activity was reported only in 22 patients who were carriers of a *BRCA1* or *BRCA2* mutation,[51] all of whom had ovarian, breast, or prostate cancer and were refractory to multiple treatment regimens.

The development of PARP inhibitors clearly does not follow the classic paradigm for cancer drug development, and novel strategies are actively being explored in both preclinical and clinical studies. This approach in part is due to the fact that the potential applications for these drugs range from cardiovascular diseases to cancer, and from conditions such as acute post-ictus recovery to chemoprevention of cancer in BRCA-mutation carriers. In the context of PARP inhibitors as anticancer agents, one would expect a high level of synergy with DNA-damaging agents, such as cytotoxic chemotherapy and radiation therapy.

The first clinical trials of a PARP inhibitor in combination with chemotherapy were performed in patients with metastatic melanoma. Combination therapy of temozolomide (200 mg/m^2) and AG014699 (Pfizer; 12 mg/m^2) in a phase II study yielded encouraging response rates (GenBank), although the hematologic toxicity of temozolomide was exacerbated.[52] There was 1 toxic death, 3 neutropenic hospitalizations, and temozolomide dose reductions in 12 of 40 patients. A large, randomized phase II trial is currently exploring temozolomide in combination with ABT-888 in melanoma. Alternative schedules of temozolomide are also available, and extended schedule regimens that cause much less neutropenia and thrombocytopenia than the 5-day regimen may offer additional opportunities to improve on this combination.

ABT-888 was evaluated in a highly innovative, first-ever phase 0 trial, which was essentially designed as a single-dose, nontherapeutic, biomarker development trial.[53] The primary goal was to validate PARP inhibition clinically in both tumor tissue and PBMCs. The initial dose of 10 mg exceeded the targeted C_{max} in the first cohort, and a trend towards inhibition of PARP activity in PBMCs as measured by an enzyme-linked immunosorbent assay was observed. Significant inhibition of PAR levels (>85% reduction) was observed in both PBMCs and in tumor

biopsies at the next dose level (25 mg). Subsequently, several phase Ib clinical trials of ABT-888 in combination with chemotherapeutic agents (temozolomide, carboplatin/paclitaxel, topotecan, and irinotecan) were initiated, thereby circumventing the traditional testing of the drug as a single agent in a classic phase I trial.[53,54]

A more conservative approach was used in the development of BSI-201 (Sanofi-Aventis), with the first phase I trial conducted as a single agent.[55] Phase Ib trials combining BSI-201 with gemcitabine, topotecan, temozolomide, and carboplatin/taxol were then initiated following the elucidation of biologically relevant doses.[56] Subsequent phase II trials of each combination were started in several tumors including ovarian, breast, and lung tumors. Results of a randomized phase II trial in triple negative breast cancer showed that BSI-201 in combination with gemcitabine and carboplatin had improved clinical benefit rate (complete response + partial response + stable disease), median progression-free survival, and, most impressively, median overall survival, compared with gemcitabine and carboplatin alone.[57]

The PARP inhibitor INO-1001 (Inotek Pharmaceuticals) was tested initially in cardiovascular diseases, after which it was granted orphan drug status by the US Food and Drug Administration for the prevention of postoperative complications of aortic aneurism repair.[58] The first phase I trial in cancer was in patients with melanoma, in which INO-1001 was combined with standard doses of temozolomide.[59]

There are still no clinical data regarding the tolerability of long-term PARP inhibition, and there are concerns that sustained prolonged inhibition of DNA repair could enhance the rate of mutations and, therefore, the potential for secondary malignancies. As a result, the development of these drugs is likely to be restricted to the setting of advanced disease until more data become available.

Further development of PARP inhibitors poses significant and unique challenges. Clinical trial designs must aim at the selection of the optimal dose and schedule of the PARP inhibitor as well as the DNA damaging agent, and subsequently identify which combinations are most appropriate for efficacy testing in phase II and III trials. The determination of the optimal dose and schedule for the PARP inhibitor will be more challenging, as surrogate endpoints for improved efficacy are not available in the early development setting.

Due to the number of cellular processes affected by PARPs, several putative biomarkers have been identified that may correlate with PARP activity. Plummer and coworkers have described a method to determine PARP activity in PBMCs and tumor tissue from patients undergoing treatment with temozolomide,[60] whereas Kinders and associates have developed an immunoassay to quantitate poly(ADP-ribose) in both PBMCs as well as tumor tissue.[61] KuDOS Pharmaceuticals has reported more than 50% PARP inhibition at 40 mg/day in preliminary data from its single-agent phase I clinical trial of KU-0059436, although the method used for determining PARP inhibition was not reported.[62] Although data generated from the use of these assays will aid in determining biologically effective doses, additional clinical data will be required to correlate these results with clinical outcomes.

Targeting the Apoptotic Pathway

The intrinsic and acquired resistance of melanoma is an area of intense investigation. One of the common mechanisms underlying the resistance of melanoma to pharmacologic therapies is the development of defects in the 2 distinct cell death pathways: apoptotic and non-apoptotic.

While the role of the non-apoptotic pathway in controlling melanoma response to therapy remains to be elucidated further, apoptosis protease activating factor-1 (Apaf-1), a key regulator of mitochondrial apoptotic pathway, was demonstrated to be closely linked to melanoma progression and resistance to chemotherapy. Apaf-1 protein levels can be restored by the addition of the hypomethylating agent decitabine or the histone deacetylase inhibitor tricostatin A.[63] Thus, although many details of the complex regulation of apoptosis in melanoma are still missing, several associations are becoming clear, which form a basis for specifically targeting melanoma apoptosis resistance.

Bcl-2 appeared to be a promising target for melanoma therapy.[64] Several groups have reported Bcl-2 expression in melanoma lesions comparable to that expressed in nevi, whereas another study found decreased Bcl-2 expression in melanoma. However, in a large series of uveal melanoma, levels of Bcl-2 expression did not correlate with disease progression.[65] Oblimersen is an antisense agent targeted to mitochondrial Bcl-2. In a phase I/II clinical study, 40% of patients with advanced melanoma (expressing Bcl-2) who received oblimersen and dacarbazine exhibited decreased Bcl-2 expression, tumor cell apoptosis, and antitumor responses.[64] Results from a randomized phase III trial in 771 patients receiving a combination of dacarbazine combined with oblimersen or dacarbazine alone showed improved progression-free survival (2.6 vs 1.6 months; $P<.01$) and response rate (13.5% vs 7.5%; $P=.007$) but no statistical difference in overall survival (9.0 vs 7.8 months; $P=.077$) in the combination arm.[66] The results of this trial were rather difficult to interpret given several issues with study design and the failure to measure tumor Bcl-2 expression. A significant interaction between baseline serum lactate dehydrogenase (LDH) and treatment was noted, with a significant increase in survival in patients with a normal LDH. As a result,

subsequent ongoing phase III studies have been designed to exclude patients with elevated LDH.

While a number of anti-apoptotic proteins including Bcl-2, Bcl-xL, and X-linked inhibitor of apoptosis (XIAP), are over-expressed in melanoma and may be linked to chemotherapy resistance, there is insufficient evidence that any is key to the central control of apoptosis.[67] Hence, targeting a single pathway is likely to be ineffective.

One of the 8 proteins from the IAP family, XIAP, blocks both the endogenous (mitochondrial) and exogenous (death receptor–related) apoptosis pathways. At present, an antisense oligonucleotide directed at XIAP is in clinical evaluation.[68] A phase II trial of the survivin inhibitor YM155 (Astellas Pharma) administered as a 7-day infusion has shown some single-agent activity, and studies in combination with chemotherapy are ongoing.[69]

Conclusions

Melanoma resistance to chemotherapy is a major impediment to improving outcomes in patients with this disease. In this review, we have presented the current status of development of agents targeted towards chemotherapy resistance abrogation that have met variable levels of success. Tremendous efforts have been expanded in developing strategies to overcome chemotherapy resistance in melanoma. A deeper understanding of the DNA repair pathways, implicated as primary mechanisms of resistance, has germinated the development of several new agents targeting DNA repair. While MGMT inhibition was not met with much success, dual inhibition approaches targeted at MGMT and MMR concomitantly may allow the exploitation of its therapeutic potential. Bcl-2 inhibition was faced with a similar disappointment; however, the promising results observed in early clinical trials of PARP inhibition may give this field additional momentum and provide chemotherapy resistance abrogation with new and exciting horizons. The successful development of DNA repair inhibitors is still incumbent upon the development of appropriate biomarkers and innovative clinical trial design. Coupling biomarker development with rational combination strategies in carefully designed, translational clinical trials offers great hope for improving therapy outcomes in patients with metastatic melanoma.

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