Novel Targets in Melanoma

Intralesional and Combination Therapy to Manipulate the Immune Response

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KEYWORDS

• Metastatic melanoma • Immunotherapy • Combination therapy
• Intralesional therapy • Checkpoint blockage

KEY POINTS

- Overall, intralesional therapy shows great promise in the early stages, although Talimogene Laherparepvec is currently the only US Food and Drug Administration-approved intralesional therapy for melanoma.
- At the time of publication, PV-10 and Bacillus Calmette-Guerin have moved into phase III clinical trials.
- Novel targets for therapy are gaining momentum and being actively pursued as agents for combination therapy in conjunction with checkpoint inhibitors for human cancers, including metastatic melanoma.
- Intralesional therapies and novel drug targets are being investigated for use with the checkpoint inhibitors and BRAF/MEK inhibition for tumors refractory to monotherapy, with promising preclinical results.
- Most combinations are in phase I safety or phase II efficacy stages of testing at the time of publication.

INTRODUCTION

Clinical outcomes for metastatic melanoma have been dramatically altered by recent developments in immunotherapy with checkpoint blockade and targeted strategies such as BRAF/MEK inhibition.[1,2] However, overall response to these therapies is not uniform, the majority of patients do not respond, and clinical response can be self-limited with eventual relapse.[1] Therefore, strategies to target the individual's specific tumor and immune profile to overcome resistance and elicit response are of paramount interest.

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To this end, current directions in melanoma treatment aim to leverage a combination of therapies for tumors refractory to anti-programmed death (PD)-1 monotherapy. Such approaches augment local immune cell recruitment beyond T cells to modify the tumor microenvironment, target oncogenes and downstream signaling, or inhibit byproducts required for tumor metabolism.[3–6] Local tumor-directed strategies such as radiation and intralesional therapy as well as inhibitors designed for novel targets may amplify currently used systemic agents when used in combination. Here, we summarize new classes of agents and emerging multimodal combination strategies that demonstrate significant promise in future melanoma management.

INTRALESIONAL THERAPIES

Talimogene iaherparepvec (T-VEC) is an oncolytic virus that mediates tumor regression through selective replication within and lysis of tumor cells as well as induction of systemic antitumor immunity capable of eradicating tumor at distant, uninjected sites. T-VEC is derived from herpes simplex virus type I and genetically modified to preferentially replicate in tumor cells and enhance immunity by increased (1) antigen loading of major histocompatibility class I molecules and (2) expression of granulocyte macrophage colony-stimulating factor to increase tumor-antigen presentation by dendritic cells.[3,7] Intratumoral T-VEC has been shown to be safe and is approved as intralesional therapy for patients with unresectable stage IIIB through IV melanoma.[8,9] Significant overall response rate (64% in injected lesions) and bystander effect (34% in uninjected lesions) was observed in phase III studies with minimal side effect profile.[7,10]

Mechanistically, T-VEC has been shown to increase the $CD8^+$ T-cell infiltrate into the tumor bed, thus priming the tumor microenvironment for treatment with checkpoint inhibitors.[11] Results from a 2-year phase II trial of T-VEC as neoadjuvant therapy in resectable stage IIIB-IVM1a melanoma (NCT02211131) also demonstrate increased $CD8^+$ cell density, which was correlated with clinical outcomes.[12] The 2- year recurrence-free survival and overall survival rates were both improved with neoadjuvant intralesional T-VEC plus surgery when compared with surgery alone.[12] T-VEC also decreases immunosuppressing regulatory T cell population and may generate long-lasting immunity, leading to durable control at uninjected sites.[3,7,11,12]

A related oncolytic virus is canerpaturev, also known as C-VEC and formerly HF10. This virus is a spontaneous mutant of herpes simplex virus 1 that preferentially replicates in tumor cells, like T-VEC.[13,14] herpes simplex virus is a focus of oncolytic research owing to its large genomic size, making it able to accommodate large transgenes, and its ability to infect a range of hosts, including common preclinical species models such as mice and monkeys.[14] Canerpaturev induces tumor cell necrosis with the increased infiltration of $CD8^+$ T cells. Studies in a mouse model have demonstrated a release of IL-2, IL-12, interferon (IFN)-alpha, IFN-beta, IFN-gamma, and tumor necrosis facto-alpha by splenocytes when exposed to squamous cell carcinoma cells in vitro following treatment with canerpaturev. This drug has been used in phase Ib trials to treat melanoma in addition to other cutaneous head and neck cancers with good clinical success (NCT01017185).[14,15]

CAVATAK is a coxsackievirus A21-based oncolytic viral therapy also used for the intralesional treatment of melanoma.[16] This virus is very common RNA virus targeting intracellular adhesion molecule-1 (ICAM-1), which is upregulated on the surface of many cancers, including melanoma.[16] Once taken in by ICAM-1–expressing cells, the cancer cell machinery is hijacked for viral replication and eventual cell lysis. The phase II study for CAVATAK demonstrated durable response of 38.6% of patients

in both injected and uninjected lesions, thereby producing the bystander effect seen in other regional therapies.[17] Follow-up extension studies showed an influx of immune cells to the tumor bed after CAVATAK injection. Additionally, digital RNA counting identified an increase in targetable checkpoint molecules, a promising finding to support combination therapy with checkpoint inhibitors.[17]

Another intralesional agent is rose Bengal disodium, a xanthene dye used in diagnostic ophthalmology and to study liver function tests.[18,19] When formulated as a 10% sterile, nonpyrogenic saline solution, it is known as PV-10, and this agent has become a focus for intralesional melanoma therapy. During preclinical testing, it was noted that lesions injected with PV-10 contained increased amounts of tumor infiltrating lymphocytes.[19] Although its precise mechanism of action is unknown, PV-10 seems to induce a systemic immune response as evidenced by local tumor destruction via lysis of tumor cells, as well as a bystander effect on uninjected lesions.[19] Phase II trials demonstrated good local control with a durable effect and complete response in 50% of patients.[18] Again, low toxicity was seen with the predominant adverse effects consisting of blistering at the treatment site. A recent propensity score-matched study demonstrated comparable survival after isolated limb infusion versus PV-10.[20]

One of the first agents to be used for intralesional injection was IL-2.[21] Initial phase II trials reported a 62.5% response rate when injected into metastatic lesions. Previously, systemic IL-2 had been used to treat metastatic melanoma, but this treatment carried significant adverse effects and toxicities.[22] More recently, intralesional IL-2 has been used in combination with topical imiquimod, because this combination is thought to restore the Th1 response (IL-2, IFN-γ, tumor necrosis factor-β) in patients with metastatic cancer, which have a predominantly Th2 response (IL-4, IL-5, IL-6, IL-10, and IL-13).[23,24] Imiquimod activates toll-like receptor 7, increasing natural killer cell activity and resulting in a robust release of cytokines to include those in the Th1 response. In clinical trials, the combination of intralesional IL-2 plus imiquimod resulted in good local control and evidence of systemic immune response, but without the toxicity of intravenous IL-2. Patients treated in this group experienced 100% clinical response rate and have demonstrated no relapse in longitudinal follow-up.[23] Ongoing trials are exploring the use of intralesional IL-2 as a fusion protein with L19, a fully human recombinant monoclonal antibody (NCT02076633, NCT01253096) and in liposomal form (NCT00004104).

A comprehensive listing of the ongoing intralesional clinical trials (early phase through phase III) for metastatic melanoma is listed in **Table 1**. Overall, intralesional therapy shows great promise in the early stages, although T-VEC is currently the only intralesional therapy approved by the US Food and Drug Administration for melanoma. At the time of publication, PV-10 and BCG have moved into phase III clinical trials.

NEW TARGETED AGENTS TO AUGMENT IMMUNOTHERAPY

An emerging group of immunotherapeutic targets present opportunities for targets for patients without BRAF mutation, or those whose tumors have developed BRAF resistance. The properties of these agents may also open the door to increased efficacy of checkpoint inhibition when used in combination to prime the immune system. These new potential targets are being studied in the context of combination therapy to complement BRAF/MEK or PD-1/PD- ligand 1 (L1) as a multifaceted manipulation of the immune system to fight metastatic disease.[1,4]

The TYRO3, AXL, and MERTK receptor tyrosine kinase family have been associated with a number of human cancers, including melanoma. Effects attributed to oncogenesis and metastasis (epithelial-to-mesenchymal transition) of the TYRO3, AXL, and

Table 1
Clinical trials of intralesional therapies for melanoma

NCT Number	Phase	Title	Interventions
NCT01209676	Early phase I	IMCgp100 in Advanced Unresectable Melanoma	IMCgp100 injection
NCT03655756	Early phase I	pDNA Intralesional Cancer Vaccine for Cutaneous Melanoma	IFx-Hu2.0
NCT01017185	Phase I	Study of HF10 in Patients With Refractory Head and Neck Cancer or Solid Tumors With Cutaneous and/or Superficial Lesions	HF10
NCT00219843	Phase I	Intralesional PV-10 Chemoablation of Metastatic Melanoma	PV-10 (rose Bengal disodium, 10%)
NCT01838200	Phase I	Phase I Study of Intralesional Bacillus Calmette-Guerin (BCG) Followed by Ipilimumab in Advanced Metastatic Melanoma	Bacillus Calmette-Guerin vaccine, ipilimumab, isoniazid
NCT03989895	Phase I	Arm 2: Intratumoral Injection of Dengue Virus-1 #45AZ5 (PV-001-DV) in Patients With Advanced Melanoma	Dengue virus-1 #45AZ5 (PV-001-DV)
NCT00815607	Phase I	Trial of an Intratumoral Injections of INXN-3001 in Subjects With Stage III or IV Melanoma	INXN-3001
NCT03747744	Phase I	Intratumoral Injection of Autologous CD1c (BDCA-1)+ Myeloid Dendritic Cells Plus Talimogene Laherparepvec (T-VEC)	CD1c (BDCA-1)+ myeloid DC + T-VEC
NCT03052205	Phase I	A Study of Intratumoral IMO-2125 in Patients With Refractory Solid Tumors	IMO-2125
NCT01123304	Phase I	Safety Study of Human IgM (MORAb-028) to Treat Metastatic Melanoma	MORAb028
NCT03803397	Phase I	Arm 1: Infusion of Autologous Monocyte-derived Lysate Pulsed Dendritic Cells (PV-001-DC) in Patients With Advanced Melanoma	PV-001-DC
NCT00003556	Phase I	Vaccine Therapy in Treating Patients With Melanoma	ALVAC-hB7.1, canarypox-hIL-12 melanoma vaccine
NCT01469455	Phase I	DNA Repair Inhibitor & Irradiation on Melanoma	DT01

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Table 1
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NCT Number	Phase	Title	Interventions
NCT00574977	Phase I	Safety Study of Modified Vaccinia Virus to Cancer	Vaccinia virus (vvDD-CDSR)
NCT02680184	Phase I	Clinical Study of CMP-001 in Combination With Pembrolizumab or as a Monotherapy	CMP-001, pembrolizumab
NCT02668770	Phase I	Ipilimumab (Immunotherapy) and MGN1703 (TLR Agonist) in Patients With Advanced Solid Malignancies	MGN1703, ipilimumab
NCT00625456	Phase I	Safety Study of Recombinant Vaccinia Virus to Treat Refractory Solid Tumors	RAC VAC GM-CSF (JX-594)
NCT01935453	Phase I	A Phase I Study of Recombinant hGM-CSF Herpes Simplex Virus to Treat Cancer	Recombinant hGM-CSF herpes simplex virus injection
NCT01986426	Phase I	LTX-315 in Patients With Transdermally Accessible Tumors as Monotherapy or Combination With Ipilimumab or Pembrolizumab	LTX-315 + ipilimumab, LTX-315 + pembrolizumab
NCT02576665	Phase I	A Study of Toca 511, a Retroviral Replicating Vector, Combined With Toca FC in Patients With Solid Tumors or Lymphoma (Toca 6)	Toca 511, Toca FC
NCT02428036	Phase I	A Study of TBI-1401(HF10) in Patients With Solid Tumors With Superficial Lesions	TBI-1401 (HF10)
NCT03301896	Phase I	Study of the Safety and Efficacy of LHC165 Single Agent and in Combination With PDR001 in Patients With Advanced Malignancies	LHC165, PDR001
NCT02988960	Phase I	A Study of ABBV-927 and ABBV-181, an Immunotherapy, in Subjects With Advanced Solid Tumors	ABBV-927, ABBV-181
NCT00022568	Phase I	Vaccine Therapy in Treating Patients With Metastatic Melanoma	Recombinant vaccinia-TRICOM vaccine

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Table 1
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NCT Number	Phase	Title	Interventions
NCT00004148	Phase I	Vaccine Therapy in Treating Patients With Unresectable Metastatic Melanoma	Recombinant vaccinia-B7.1 vaccine
NCT00005057	Phase I	Gene Therapy and Ganciclovir in Treating Patients With Stage IV Melanoma	Adenovirus RSV-TK, ganciclovir
NCT03712358	Phase I	PVSRIPO for Patients With Unresectable Melanoma	PVSRIPO
NCT00623831	Phase I	A Phase 1 Study of Mixed Bacteria Vaccine (MBV) in Patients With Tumors Expressing NY-ESO-1 Antigen	Mixed bacterial vaccine
NCT01082887	Phase I Phase II	A Study of Immunotherapy With TIL (Tumor Infiltrating Lymphocytes) in Combination With Intra-tumoral Injections of Interferon Gamma-adenovirus (Ad-IFNg) in Patients With Stage IIIc or Stage IV Metastatic Melanoma (AJCC)(Protocol TIL-Ad-INFg)	TIL + INFG
NCT02225366	Phase I Phase II	Intratumoral Injections of LL37 for Melanoma	LL37
NCT01397708	Phase I Phase II	Safety Study of Adenovirus Vector Engineered to Express hIL-12 in Combination With Activator Ligand to Treat Melanoma	INXN-2001
NCT00429312	Phase I Phase II	A Study of Recombinant Vaccinia Virus to Treat Malignant Melanoma	JX-594
NCT00002817	Phase I Phase II	Vaccine Therapy in Treating Patients With Metastatic Melanoma	Sargramostim, vaccinia-GM-CSF vaccine
NCT01227551	Phase II	A Study of Intratumoral CAVATAK in Patients With Stage IIIc and Stage IV Malignant Melanoma (VLA-007 CALM)	Coxsackievirus A21 (CVA21, CAVATAK)
NCT01253096	Phase II	Intratumoral Application of L19IL2 in Patients With Malignant Melanoma	L19IL2

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NCT Number	Phase	Title	Interventions
NCT01636882	Phase II	CAVATAK in Patients With Stage IIIc or IV Malignant Melanoma to Extend Dosing to 48 Weeks (VLA-008 CALM Ext)	CVA21
NCT00521053	Phase II	Phase 2 Study of Intralesional PV-10 for Metastatic Melanoma	PV-10
NCT00204581	Phase II	Intralesional Treatment With Interleukin-2 (Proleukin) in Soft Tissue Melanoma Metastases	IL-2
NCT02076633	Phase II	Intratumoral Administration of L19IL2/ L19TNF	L19IL2 + L19TNF
NCT03190824	Phase II	Evaluate Efficacy, Immunologic Response of Intratumoral/ Intralesional Oncolytic Virus (OBP-301) in Metastatic Melanoma	OBP-301
NCT03754140	Phase II	Intralesional Sclerosant for in Transit and Cutaneous Melanoma Metastases	Polidocanol injection
NCT03928275	Phase II Phase III	The Response to Intralesional IL-2 and/or BCG Treatment for Cutaneous Metastatic Melanoma	Bacillus Calmette-Guerin + IL-2
NCT02288897	Phase III	PV-10 vs Chemotherapy or Oncolytic Viral Therapy for Treatment of Locally Advanced Cutaneous Melanoma	PV-10 + dacarbazine, temozolomide or talimogene laherparepvec

MERTK receptors have been described. In particular, the AXL tyrosine kinase receptor has been implicated with poorer prognosis in many human cancers, including certain resistant phenotypes of melanoma and failure to respond to targeted therapy or checkpoint blockade.[25–27] Similar to immune checkpoint, AXL serves as a negative immune regulator in response to inflammation and injury, a mechanism that is high-jacked in the tumor setting to promote unchecked growth.[25–30] AXL receptor function significantly manipulates epithelial to mesenchymal plasticity in the tumor microenvironment, influencing ligand secretion by macrophages. This finding has been correlated in preclinical in vivo studies with significant increase in metastatic potential for AXL-expressing tumors, with metastatic events eliminated by multiple strategies of AXL inhibition. Response was greatest in immunocompetent models, highlighting the interplay between this pathway and immune cell function.[31] The interaction of AXL with various cellular process is illustrated in **Fig. 1**.[27]

The AXL pathway may critically contribute to adaptive or evasive immune resistance because it is seen to be significantly activated in chemotherapy resistant breast and

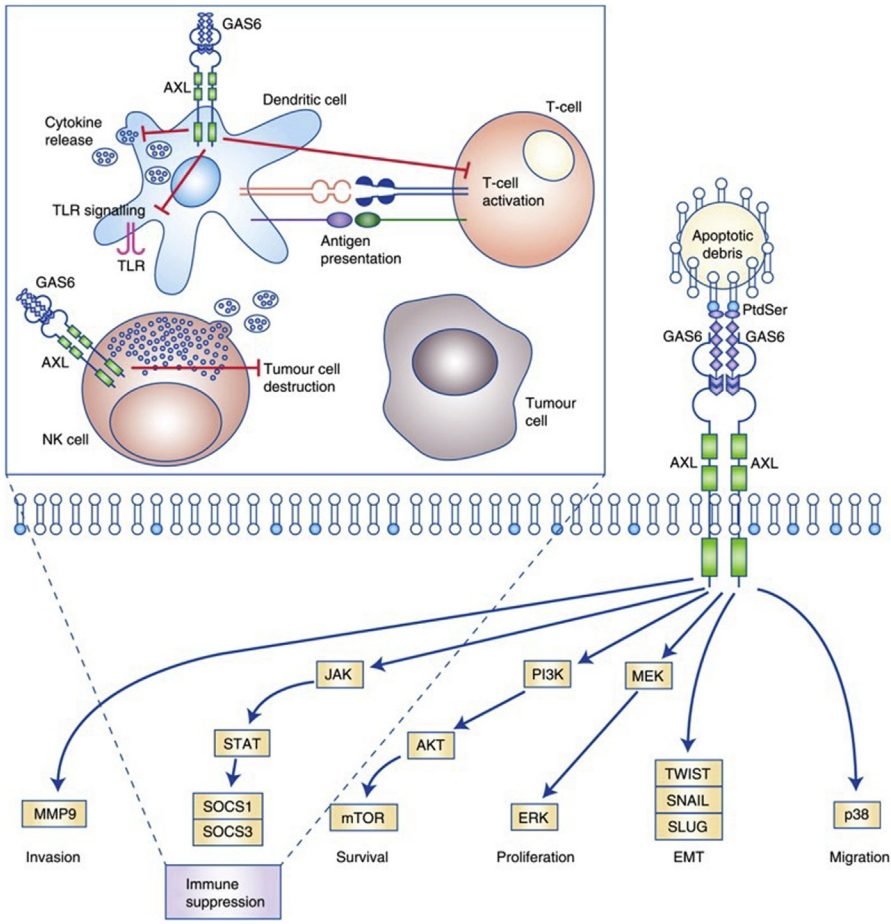

Fig. 1. Spectrum of cellular processes regulated by AXL activity. AXL, after activation by its ligand growth arrest-specific 6 along with an interaction between growth arrest-specific 6 and phosphatidylserine (PtdSer), dimerises and cross-phosphorylates (*yellow circle*) its partner receptor. This activation regulates an array of cellular pathways as illustrated at the bottom of the figure. *Inset*: AXL activity plays a complex role in immune regulation that includes the inhibition of cytokine release, toll-like receptor signaling, and T-cell activation by antigen-presenting cells such as dendritic cells (above), as well as specific antitumor killing by natural killer cells (below). (*From* Gay, C.M., K. Balaji, and L.A. Byers, *Giving AXL the axe: targeting AXL in human malignancy.* British Journal of Cancer, 2017. 116(4): p. 415; with permission.)

pancreatic models as well as radioinsensitive melanoma.[25,27] Early preclinical data demonstrates Axl ligand, growth arrest-specific 6 influences suppressive function of regulatory T cells and inhibits natural killer and dendritic cell maturation, thereby dampening antitumor immune response.[28,30,32,33] Axl expression has been shown to predict checkpoint blockage failure in melanoma patients.[26] Preclinical models investigating Axl-inhibition in combination with PD-1 demonstrated a synergistic effect evidenced by unregulated PD-L1 expression.[31] Mechanistic studies unveiled that Axl inhibition increased the proliferation of $CD8^+$ T cells, likely through dendritic cells and the activation of $CD103^+$ during cross-presentation.[31] Anti-Axl is being investigated

with the drug TP-0903 (NCT02729298) to treat all solid tumors, including melanoma. A second anti-Axl small molecule inhibitor, bemcentinib, has recently shown efficacy in a phase II trial (NCT03184571) for lung adenocarcinoma in patients with low/no PD-1 expression. Moreover, the combination of bemcentinib and pembrolizumab was well-tolerated.[34]

Indoleamine 2,3-dioxygenase (IDO), an immunomodulatory enzyme is produced by myeloid-derived suppressor cells and inhibits the immune response by depletion of the amino acid tryptophan, thereby affecting tumor metabolism and altering the tumor microenvironment type.[35] IDO contributes to immune suppression and tolerance by suppressing the proliferation and differentiation of effector T cells, and enhancing the activity of regulatory T cells.[36,37] In the absence of tumor, IDO is systemically required for tolerance. This enzyme is found in the gut and on mucosal surfaces, and IDO-deficient mice have been shown to reject their fetuses. IDO works by direct suppression of proinflammatory TH17 cytokines IL-6 and IL-17. However, in the setting of cancer, this natural promotion of immune tolerance is hijacked by tumors and leads to unchecked tumor growth. In immunogenically cold lesions, the inhibition of IDO could prevent immune tolerance and allow for robust immune response when followed by the administration of intralesional oncolytic virus.[36,37] In a companion canine model of metastatic melanoma and sarcoma, systemic IDO inhibition was found to augment response to local treatment with radiotherapy with good efficacy and limited toxicity.[38] Anti-IDO is being investigated as DX-03-12 and DX-03-13 (NCT03047928) used in combination nivolumab and PD-L1/IDO peptide vaccine. A separate trial, NCT04007588, uses BMS-986205 as the anti-IDO drug in the presence and absence of both nivolumab and ipilimumab.

Efficiency of the T-cell response is paramount to harnessing the immune system for tumor eradication. Certain T-cell markers, among them TIGIT and LAG3, have been found to represent an exhausted phenotype of T cells, characterized by decreased cytotoxicity and effector function.[39,40] Lymphocyte activating gene, in particular, is expressed on $CD4^+$ and $CD8^+$ T cells and has been shown to act in synergy with PD-1 to downregulate T cells. It is thought that the lymphocyte activating gene operates through preventing calcium influx via T-cell receptors, which downstream leads to decreased expansion and eventual reduction in the amount of memory T cells. Evidence of enriched coexpression of lymphocyte activating gene and PD-1 was seen on tumor infiltrating lymphocytes in ovarian cancer.[40] A second method to circumvent T-cell exhaustion lies in the interaction between T-cell Ig and ITIM domain (TIGIT), which is expressed by tumor infiltrating lymphocytes, and CD155, which is expressed by tumor cells, and in particular melanomas.[39] TIGIT expression is noted to be highest on $CD8^+$ T cells. When bound by the inhibitory CD155, this induces inhibition of the classic $CD8^+$ T-cell effector function and allows the tumor to evade immune response.[39] Anti-LAG3 is being investigated as adjuvant therapy with nivolumab and pembrolizumab (NCT02676869, NCT03743766) and in a separate trial with nivolumab and ipilimumab (NCT02519322). The anti-TIGIT drug BGB-A1217 has an ongoing study with tislelizumab (and anti-PD1 agent) in advanced solid tumors (NCT04047862). A recent study of anti-TIGIT plus nivolumab was terminated (NCT03119428).

INTRALESIONAL AND SYSTEMIC COMBINATION THERAPIES

As is well-known, the response rate for checkpoint inhibition and intralesional therapy is not universal. Although it has long been accepted that a tumor's genetic mutational burden drives its biologic behavior, there is a growing appreciation for the role of the

immune phenotype of the tumor to predict and dictate response to treatment, particularly immunotherapy. Hot tumors are have high immunologic infiltrate, whereas cold tumors are non–T-cell inflamed cancers.[41,42] Hot tumors have higher expression of T-cell markers, IFN, IDO, and PD-1, to name a few, and thus are more responsive to immunotherapy. Naturally, the next steps involve questions of how we may manipulate the immune phenotype of tumors to elicit a greater pathologic response. To this end, combination therapy leveraging intralesional therapies and systemic therapies is being explored (**Table 2**).

The combination of T-VEC plus anti–PD-1 therapy was first investigated by Ribas, and colleagues[11] Intratumoral injection with T-VEC resulted in an influx of $CD8^+$ T cells into the tumor, increased PD-L1 protein expression, and induced IFN-γ gene expression. When T-VEC was administered in combination with ipilimumab, a synergistic effect was seen as evidenced by improved efficacy. Importantly, as seen in their phase Ib trial, this effect did not occur in the setting of increased toxicity.[8,11] The phase III follow-on study, MASTERKEY-265, is currently active with 713 participants to evaluate T-VEC plus pembrolizumab when compared with placebo injection plus pembrolizumab for 24 months from the date of first treatment or disappearance of lesions (NCT02263508).

Canerpaturev (HF10) has been tested in phase II trials in combination with ipilimumab for patients with unresectable/unresected stage IIIB to IV melanoma (NCT02272855). This combination demonstrated a median progression-free survival of 19 months and a median overall survival of 26 months.[43,44] Molecular studies of the tumors revealed increased $CD8^+$ T-cell infiltrate and decreased $CD4^+$ T cells.[43] In this study, 28.3% of patients had grade 3 or higher adverse events, most which were attributed to ipilimumab and only 3 attributed to intralesional therapy.[43]

Combination therapy has been studied using CAVATAK and PD-1 blockade in an immune competent murine model using B16-ICAM-1 melanoma.[45] Authors in this study noted greater antitumor activity of the CAVATAK plus PD-1 monoclonal antibody when compared with saline controls. Although this method does rely on the expression of ICAM-I by the tumor cells, a survival benefit was seen in the mice. Based on this preclinical evidence, phase I trials combining CAVATAK with either pembrolizumab (NCT02565992) or ipilimumab (NCT02307149) have been initiated in the United States.[46] A second preclinical trial in immune competent mice was conducted to study CAVATAK in addition to PD-1 blockade and anti-IDO. CAVATAK plus anti-IDO alone was not associated with a significant reduction in tumor size, but triple therapy was superior to CAVATAK plus PD-1 blockade alone.[47]

A completed phase I trial examined the response of intratumoral IL-2 and intratumoral ipilimumab in 12 patients (NCT01672450). Because the systemic checkpoint inhibitors are not without side effects, it was postulated that intralesional injections of each agent would decrease systemic toxicity. Overall, the local response to therapy was 67% and a bystander response was seen in 89%.[48] There were no dose-limiting toxicities and all patients completed treatment. A separate phase II trial examined intralesional IL-2 with systemic ipilimumab (NCT01480323). The overall efficacy rate in this study was 20%, with 40% of patients experiencing a grade III/IV adverse event.[49] The results ultimately suggested that the combination therapy did not show improved efficacy over ipilimumab alone.

An international phase Ib/II study is currently underway to determine the safety and efficacy of PV-10 in combination with pembrolizumab (NCT02557321). Patients with metastatic melanoma will receive either PV-10 plus pembrolizumab or pembrolizumab alone. There are no data available as of yet, because the primary study completion date is anticipated to occur in April 2020. Before this trial, a small, 3-person case series

Table 2
Clinical trials of combinations with intralesional and checkpoint therapies

NCT Number	Phase	Title	Interventions
NCT01672450	Phase I	A Study of Intratumoral Injection of Interleukin-2 and Ipilimumab in Patients With Unresectable Stages III-IV Melanoma	Intratumoral ipilimumab and IL-2
NCT02565992	Phase I	Intratumoral CAVATAK (CVA21) and Pembrolizumab in Patients With Advanced Melanoma (VLA-011 CAPRA)	CAVATAK + pembrolizumab
NCT02307149	Phase I	Intratumoral CAVATAK (CVA21) and Ipilimumab in Patients With Advanced Melanoma (VLA-013 MITCI)	CAVATAK + ipilimumab
NCT03291002	Phase I	Study of Intratumoral CV8102 in cMEL, cSCC, hnSCC, and ACC	CV8102 + anti-PD-1 therapy
NCT03773744	Phase I	MG1-MAGEA3 With Ad-MAGEA3 and Pembrolizumab in Patients With Previously Treated Metastatic Melanoma or Cutaneous Squamous Cell Carcinoma	Ad-MAGEA3, MG1-MAGEA3, pembrolizumab, cyclophosphamide
NCT03084640	Phase I	Phase 1B Study Evaluating Alternative Routes of Administration of CMP-001 in Combination With Pembrolizumab in Participants With Advanced Melanoma	CMP-001, pembrolizumab
NCT02680184	Phase I	Clinical Study of CMP-001 in Combination With Pembrolizumab or as a Monotherapy	CMP-001, pembrolizumab
NCT02668770	Phase I	Ipilimumab (Immunotherapy) and MGN1703 (TLR Agonist) in Patients With Advanced Solid Malignancies	MGN1703, ipilimumab
NCT01986426	Phase I	LTX-315 in Patients With Transdermally Accessible Tumors as Monotherapy or Combination With Ipilimumab or Pembrolizumab	LTX-315 + ipilimumab, LTX-315 + pembrolizumab
NCT01838200	Phase I	Phase I Study of Intralesional Bacillus Calmette-Guerin (BCG) Followed by Ipilimumab in Advanced Metastatic Melanoma	Bacillus Calmette-Guerin vaccine, ipilimumab, Isoniazid

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NCT Number	Phase	Title	Interventions
NCT02557321	Phase I Phase II	PV-10 in Combination With Pembrolizumab for Treatment of Metastatic Melanoma	PV-10, pembrolizumab
NCT03474497	Phase I Phase II	UCDCC#272: IL-2, Radiotherapy, and Pembrolizumab in Patients Refractory to Checkpoint Blockade	IL-2, pembrolizumab, radiotherapy
NCT02719015	Phase I Phase II	Dose Escalation and Cohort Expansion of Safety and Tolerability Study of Intratumoral rAd.CD40L (ISF35) in Combination of Systemic Pembrolizumab in Patients With Refractory Metastatic Melanoma	rAd.CD40L, pembrolizumab
NCT02644967	Phase I Phase II	A Study to Assess the Safety and Efficacy of Intratumoral IMO-2125 in Combination With Ipilimumab or Pembrolizumab in Patients With Metastatic Melanoma	IMO-2125, ipilimumab, pembrolizumab
NCT02521870	Phase I Phase II	A Trial of Intratumoral Injections of SD-101 in Combination With Pembrolizumab in Patients With Metastatic Melanoma or Recurrent or Metastatic Head and Neck Squamous Cell Carcinoma	SD-101(1), pembrolizumab
NCT03958383	Phase I Phase II	IT-hu14.18-IL2 With Radiation, Nivolumab and Ipilimumab for Melanoma	hu14.18-IL2, radiation, nivolumab, ipilimumab
NCT02706353	Phase I Phase II	APX005M in Combination With Systemic Pembrolizumab in Patients With Metastatic Melanoma	APX005M, pembrolizumab
NCT03684785	Phase I Phase II	Intratumoral AST-008 Combined With Pembrolizumab in Patients With Advanced Solid Tumors	AST-008 + pembrolizumab
NCT03058289	Phase I Phase II	A Phase 1/2 Safety Study of Intratumorally Dosed INT230–6	INT230–6, anti–PD-1 antibody, anti–CTLA-4 antibody
NCT03767348	Phase I Phase II	Study of RP1 Monotherapy and RP1 in Combination With Nivolumab	RP1, nivolumab

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NCT Number	Phase	Title	Interventions
NCT03435640	Phase I Phase II	A Study of NKTR-262 in Combination With NKTR-214 and With NKTR-214 Plus Nivolumab in Patients With Locally Advanced or Metastatic Solid Tumor Malignancies	NKTR-262, bempegaldesleukin, nivolumab
NCT03058289	Phase I Phase II	A Phase 1/2 Safety Study of Intratumorally Dosed INT230–6	INT230-6, anti–PD-1 antibody, anti–CTLA-4 antibody
NCT01480323	Phase II	A Phase II Study to Evaluate Safety and Efficacy of Combined Treatment With Ipilimumab and Intratumoral Interleukin-2	IL-2 + ipilimumab
NCT03259425	Phase II	Neoadjuvant Trial of Nivolumab in Combination With HF10 Oncolytic Viral Therapy in Resectable Stage IIIB, IIIC, IVM1a Melanoma	Nivolumab, HF10
NCT02272855	Phase II	A Study of Combination Treatment With HF10 and Ipilimumab in Patients With Unresectable or Metastatic Melanoma	HF10, ipilimumab
NCT03132675	Phase II	Tavo and Pembrolizumab in Patients With Stage III/IV Melanoma Progressing on Pembrolizumab or Nivolumab Treatment	Tavokinogene telseplasmid, pembrolizumab
NCT02493361	Phase II	Trial of pIL-12/MK-3475 in Metastatic Melanoma	Pembrolizumab + pIL-12
NCT03153085	Phase II	A Study of Combination With TBI-1401(HF10) and Ipilimumab in Japanese Patients With Unresectable or Metastatic Melanoma	TBI-1401(HF10), ipilimumab
NCT03618641	Phase II	CMP-001 in Combo With Nivolumab in Stage IIIB/C/D Melanoma Patients With Clinically Apparent Lymph Node Disease	CMP-001, nivolumab
NCT04093323	Phase II	Polarized Dendritic Cell (aDC1) Vaccine, Interferon Alpha-2, Rintalolimid, and Celecoxib for the Treatment of HLA-A2+ Refractory Melanoma	Alpha-type-1 polarized dendritic cells, celecoxib, PD-1 ligand inhibitor, PD1 inhibitor, recombinant interferon alfa-2b, rintatolimod

(*continued on next page*)

Table 2 (*continued*)

NCT Number	Phase	Title	Interventions
NCT03445533	Phase III	A Study of IMO-2125 in Combination With Ipilimumab vs Ipilimumab Alone in Subjects With Anti-PD-1 Refractory Melanoma (ILLUMINATE-301)	Ipilimumab, IMO-2125

was performed to describe the effects of PV-10 plus radiation therapy to the injected lesion.[50] This regimen was well-tolerated with no significant adverse events, but not validated in a follow-on controlled trial.

SUMMARY

In summary, research surrounding intralesional therapy, novel molecular targets and combination therapy is a dynamic and promising direction for melanoma research based on preclinical and early clinical evidence. In the near future, we may see utilization of such agents and combinations more readily in the clinical setting, with the potential to benefit a larger scope of patients beyond current available therapies. These trials are actively changing the landscape of surgical approach to locally advanced and metastatic melanoma and will be important in the surgical armamentarium as neoadjuvant therapies become more standard.

DISCLOSURE

The authors of this article have no commercial or financial interests to disclose.

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