Mechanism of Action of Immunotherapy

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The immune system plays a vital role in regulating the growth of tumors. Some types of inflammatory responses can promote tumor growth, while a tumor-specific adaptive immune response can potentially control tumor growth. Malignancies have the ability to evade the immune system, and proliferate and metastasize. The goal of immunotherapy is to marshal the specificity and long-term memory of the adaptive immune response to achieve durable tumor regression and possible cure, although, to date, this has been achieved in only a small subset of patients. A variety of approaches to immunotherapy have been investigated. These include administration of exogenous cytokines or therapeutic vaccines to increase the frequency of tumor-specific T cells, adoptive transfer of tumor-specific immune effector cells, and, more recently, the application of a variety of immune checkpoint inhibitors and agonists of co-stimulatory receptors to overcome tumor-induced immune-suppressive mechanisms. Some approaches have been more successful than others for reasons that are now becoming apparent, and these observations have led to an exciting resurgence in clinical research to develop more effective immunotherapeutic strategies.
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INTRODUCTION AND RATIONALE FOR IMMUNOTHERAPY

It has long been recognized that the immune system and malignant cells often coexist in a dynamic equilibrium, and the complex interaction between growing tumors and the immune system may determine the course of disease.[1–3] Tumors must develop the ability to evade the immune system in order to proliferate and metastasize. The theory of immune surveillance suggests that the immune system is proactively able to eliminate abnormal cells and prevent cancer formation in the body. Studies have shown that patients with compromised or suppressed immune function are at increased risk of developing cancer.[4–10] Additionally, although controversial, use of immunosuppressive agents has been associated with an increased incidence of certain cancers.[11] Clearly the adaptive immune response is able to control the growth of some tumors, as evidenced by the observation that the presence of tumor-infiltrating lymphocytes (TILs) often is associated with improved overall survival (OS).[12–14] However, the immune system is rendered ineffective as tumors progress. The dynamic process of cancer immunoediting can be conceptualized by a seesaw that balances immune protection with immune evasion.[15] Cancer immunoediting involves three stages: elimination, equilibrium, and escape. In the elimination stage, cancer cells are identified and effectively destroyed by the immune system. In the equilibrium stage, the immune system is unable to completely eliminate all cancer cells but is able to control or prevent further outgrowth. The conceptual seesaw is, therefore, balanced in the equilibrium stage. In the escape stage, the immune system is unable to eliminate and control the outgrowth of the tumor because the cancer cells have evolved under the selective pressure of the immune system, and those cells that have acquired the ability to suppress or evade the immune response continue to proliferate and spread.

The goal of cancer immunotherapy is to boost or restore the ability of the immune system to detect

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and destroy cancer cells by overcoming the mechanisms by which tumors evade and suppress the immune response,[16] in essence to shift the equilibrium back in favor of immune protection. The hallmark of the adaptive immune response is specificity and long-term memory, which, when present, can result in durable responses. The traditional approach to immunotherapy has been to increase the frequency of tumor-specific T cells through administration of tumor vaccines, cytokines such as interleukin (IL)-2, and adoptive transfer of TILs. In the last decade, efforts to improve presentation of tumor antigens to the immune system have focused on antigen-presenting cells (APCs) such as myeloid dendritic cells (DCs). Another approach is to trigger innate immune activation and inflammation in the tumor microenvironment with agents such as type I interferons (IFNs) and Toll-like receptor (TLR) agonists. More recently, improved understanding of immune regulatory mechanisms, such as cytotoxic T-lymphocyte–associated antigen 4 (CTLA-4), programmed cell death 1 (PD-1), indoleamine-2,3-dioxygenase (IDO), regulatory T cells (Tregs), and myeloid-derived suppressor cells (MDSCs), has led to the development of agents that can modulate the so-called "immune checkpoints" and new strategies to deplete Tregs and MDSCs. Immune checkpoint inhibitors have generated excitement because these agents appear to overcome the very mechanisms that tumors hijack in order to suppress the antitumor immune response. Some of these modalities have been tested clinically for decades, whereas others are new, and even some of the older strategies are being revisited with new twists based on our current understanding of immuno-oncology. This article reviews the mechanism of action of each of these approaches.

CYTOKINES

Cytokines have multiple and diverse biologic activities.[17] They can be grouped into different functional classes including IFNs, ILs, chemokines, mesenchymal growth factors, tumor necrosis factors (TNFs), and adipokines. The majority of cytokines serve as ligands for specific cell-surface receptors and some act as transcription factors. The multiple and varied activities of a single cytokine can be attributed to differential expression of the cognate receptor or the specific cell type in which the cytokine is expressed.

Cytokines have been used as cancer immunotherapy for decades, and they work by one of two general mechanisms: either by exerting a direct antitumor effect or by indirectly enhancing the antitumor immune response.[18] Numerous in vitro and animal studies have shown that TNF-α and IL-6 can directly affect tumor cell growth and survival. Use of these particular direct-acting cytokines in patients with cancer, however, has proven less successful because of significant toxicity. For instance, although TNF-α and IL-6 are able to suppress growth of some tumors, they actually promote growth of others, and IL-6 can be immunosuppressive. Therefore, the use of cytokines that have a direct antitumor effect remains primarily an academic pursuit.

In contrast, cytokines that stimulate an antitumor immune response through a variety of different pathways have been widely used in clinical practice.[18] For example, IL-2 and IFN-α promote growth and activation of T cells and natural killer (NK) cells, whereas granulocyte-macrophage colony-stimulating factor (GM-CSF) acts on APCs to increase antigen processing and presentation as well as production of co-stimulatory cytokines. These cytokines are well-established cancer immunotherapies. For example, IL-2 is approved by the US Food and Drug Administration (FDA) for the treatment of metastatic melanoma and metastatic renal cell carcinoma, and IFN-α is approved for the treatment of malignant melanoma, as well as various types of leukemia and lymphoma.[19–22] Of note, GM-CSF is approved for its hematopoietic reconstitution effect and not its antitumor effect. Several other cytokines, such as IL-7, IL-11, IL-12, IL-15, IL-21, IFN-β, and IFN-γ, also are being evaluated as cancer immunotherapies.

THERAPEUTIC CANCER VACCINES

The goal of therapeutic cancer vaccines is to increase presentation of tumor-associated antigens (TAAs) to the immune system and to increase activation of tumor-specific T cells and B cells.[23] Cancer vaccines can be classified into several major categories: cell-based vaccines, protein/peptide vaccines, and genetic vaccines (Table 1).

Tumor cell vaccines can be either autologous (derived from patient-specific tumor cells) or allogeneic (produced from established human tumor cell lines).[24] The advantage of cell-based vaccine approaches lies in the potential to present the entire spectrum of tumor-associated antigens to the patient's immune system (ie, polyvalent). For autologous vaccines, tumor cells are isolated from the patient, irradiated, combined with an immunostimulatory agent, and then infused or injected back into the same patient. However, this technique requires sufficient tumor specimen and is, thus, limited to certain tumor types, and has cost and feasibility barriers. Allogeneic vaccines may overcome some of the limitations of autologous vaccines because they are derived from established tumor cell lines and can be produced on a large scale. The

Table 1. Therapeutic Cancer Vaccines in Clinical Development

Vaccine Class	Vaccine Name	Description	Clinical Development Status by Tumor Type			
			Phase I	Phase II	Phase III	Approved
Tumor cell	Algenpantucel-L (HyperAcute, NewLink Genetics, Ames, IA)	Allogeneic human pancreatic cancer vaccine based on the concept of hyperacute rejection	RCC, prostate	Melanoma, NSCLC	Pancreatic	–
	Pancreatic tumor cell vaccine (GVAX, Aduro Biotech, Berkeley, CA)	GM-CSF gene-transfected tumor cell vaccine	MM, melanoma	Pancreatic, AML, CML	–	–
	SL-701	Multivalent glioma-associated antigen vaccine	–	GBM	–	–
DC/APCs	Sipuleucel-T (Provenge Dendreon Corp, Seattle, WA)	Autologous PBMCs activated with PAP-GM-CSF	–	–	–	Prostate
	AGS-003	Autologous DCs transfected with tumor and CD40L RNAs	–	–	RCC	–
	DCVAC/Pca	Autologous DCs pulsed with killed prostate cancer line LNCap	–	Prostate	Prostate	–
	DCVax-L (Northwest Biotherapeutics, Bethesda, MD)	Autologous DCs pulsed with tumor lysate antigen	–	–	GBM	–
	CVac (Prima BioMed, Sydney, Australia)	Autologous DCs pulsed with MUC1-mannan fusion protein	–	Ovarian	–	–
	GRNVAC1	Autologous DCs transfected with hTERT and LAMP-1	–	AML	–	–
	ICT-107	Autologous DCs pulsed with antigens	–	GBM	–	–
	Ovapuldencel-T	Autologous PBMCs in GM-CSF	–	Ovarian, peritoneal	–	–
	MelCancerVac (DanDrit Biotech, Randolph, VT)	Autologous DCs pulsed with allogeneic melanoma cell lysate	–	CRC, NSCLC	–	–
	DCP-001	Allogeneic dendritic progenitor cells (DCOne™)	–	AML	–	–
	BPX-201	Autologous DCs engineered with the DeCIDe technology to target prostate cancer cells	Prostate	–	–	–

Table 1 (*continued*)

Vaccine Class	Vaccine Name	Description	Clinical Development Status by Tumor Type			
			Phase I	Phase II	Phase III	Approved
Peptides/ proteins	GV1001	hTERT peptide	Melanoma, pancreatic	HCC	NSCLC, pancreatic	–
	Nelipepimut-S (NeuVax, Galena Biopharma, Inc, Portland, OR)	HER2/*neu* peptide combined with GM-CSF	–	–	Breast	–
	L-BLP25 (Tecemotide)	Liposome-encapsulated synthetic peptide derived from MUC-1	–	Rectal, NSCLC, MM, prostate, CRC	NSCLC	–
	Rindopepimut	hEGFR variant III specific peptide conjugated to KLH	–	GBM	GBM	–
	POL-103A	Protein antigens from 3 melanoma cell lines with alum adjuvant	–	–	Melanoma	–
	IMA901	Synthetic vaccine consisting of 10 different TUMAPs	–	–	RCC	–
	MAGE-A3	MAGE-A3 combined with GM-CSF	MM	Bladder	Melanoma	–
	PVX-410	Multi-peptide vaccine	MM	–	–	–
	IMA950	Multi-peptide glioma vaccine containing TUMAPs	GBM	GBM	–	–
Genetic	Rilimogene galvacirepvec (PROSTVAC, Bavarian Nordic, Kvistgaard, Denmark)	Recombinant fowlpox/vaccinia virus encoding hPSA and TRICOM	Prostate	Prostate	Prostate	–
	CG0070	Oncolytic adenovirus encoding GM-CSF	–	Bladder	–	–
	TG4010	Modified vaccinia virus encoding human MUC1 and IL-2	Solid tumors	NSCLC	–	–

Abbreviations: AML, acute myeloid leukemia; APC, antigen-presenting cell; CML, chronic myelogenous leukemia; CRC, colorectal cancer; DC, dendritic cell; EGFR, epidermal growth factor receptor; GBM, glioblastoma; GM-CSF, granulocyte-macrophage colony-stimulating factor; HCC, hepatocellular carcinoma; hPSA, human prostate specific antigen; hTERT, human telomerase reverse transcriptase; IL-2, interleukin-2; KLH, keyhole limpet hemocyanin; MM, multiple myeloma; MUC1, mucin 1; NSCLC, non-small-cell lung cancer; PBMC, peripheral blood mononuclear cells; RCC, renal cell carcinoma; TRICOM, recombinant vaccinia virus vaccine encoding 3 co-stimulatory molecule transgenes B7.1, ICAM-1, and LFA-3; TUMAPs, tumor-associated peptides.
Note: This is not a complete list of all therapeutic cancer vaccines in clinical development.
Source: ClinicalTrials.gov.

classic example of a polyvalent, allogeneic cancer vaccine is CancerVax (CancerVax, Delaware), the whole-melanoma cell vaccine for advanced melanoma.[25] However, this vaccine was never shown to improve OS, and development was halted.[26] A more recent example that is currently in late-stage development is TRICOM, a combination of 3 immunostimulants (B7.1, ICAM-1, and LFA-3) with algenpantucel-L (HyperAcute, New Link, Ames, Iowa), a whole-cell vaccine containing irradiated

pancreatic tumor cells modified to express alpha 1,3-galactosyltransferase.

A variety of DC-based vaccines are also in development (Table 1). Dendritic cells are the most potent APCs and serve to bridge the gap between innate and adaptive immunity.[27] The main role of DCs is to uptake, process, and present pathogen-derived or host-derived antigenic peptides to naïve T cells in peripheral tissues.[28,29] Numerous studies have evaluated vaccines containing peptide-pulsed or protein-pulsed, or viral-vector infected DCs in a variety of tumor types.[30,31] Sipuleucel-T, which consists of APCs from peripheral blood mononuclear cells that have been incubated with prostatic acid phosphatase (PAP) fused to GM-CSF, was the first FDA-approved autologous cell vaccine and is indicated for men with metastatic castration-resistant prostate cancer.[32,33]

Protein/peptide-based cancer vaccines are generally derived from specific TAAs and are typically administered with an adjuvant or immune modulator to enhance the immune response to that peptide.[24] Investigators at the National Cancer Institute pioneered the identification and cloning of tumor-associated peptide antigens recognized by TILs isolated from patients.[34] A potential disadvantage of this approach is that protein/peptide-based vaccines target only one or a few epitopes of the TAA, and peptide antigens are not processed and presented by APCs in the same way that antigens from whole cells are. Therefore, they may not stimulate an effective T-cell–mediated immune response. It is generally thought that cancer vaccines must induce both antigen-specific $CD4^+$ helper T cells and $CD8^+$ cytotoxic T lymphocytes (CTLs) to be effective. Nevertheless, several protein/peptide-based vaccines are currently in clinical development. For example, DPX-0907, which was created from human leukocyte antigen (HLA)-A2 restricted peptides, has yielded encouraging results in an early trial in patients with advanced-stage breast, ovarian, or prostate cancers.[35] An epidermal growth factor (EGF)-based protein vaccine is also being investigated in patients with advanced non-small-cell lung cancer.[36] Additional peptide-based approaches using longer epitopes that will elicit both CD4 and CD8 T cells in a non–HLA-restricted fashion are currently under clinical development.

Genetic vaccines (DNA-based, RNA-based, and viral-based) can be used to induce somatic cells or DCs to express TAAs, thus resulting in cross-priming or direct antigen presentation (Table 1).[24] An important advantage of genetic vaccines is the ability to deliver multiple antigens in a single immunization, resulting in the activation of different types of immunity.[37] Initially, DNA vaccines showed promise in preclinical studies; however, clinical results have been disappointing.[38–40] Novel constructs and methods of administration are currently in development and may improve the effectiveness of DNA vaccines. Unlike DNA vaccines, RNA vaccines are rapidly degraded and cleared, and thus are less likely to cause side effects.[24] Early phase I/II trials of RNA vaccines have been conducted in patients with melanoma and renal cell carcinoma (RCC).[41–48] Viral vectors with low disease-causing potential and low intrinsic immunogenicity, such as poxvirus, adenovirus, and Herpes simplex virus type 1, have been evaluated in late-stage clinical trials.[24]

ADOPTIVE T-CELL THERAPY

Adoptive T-cell therapy involves infusion of ex vivo activated and expanded tumor-specific T cells into the patient.[49] Various sources and types of T cells have been used for adoptive therapy, including TILs, engineered T cells that express a cancer-specific T-cell receptor (TCR), and engineered T cells that express a chimeric antigen receptor (CAR). Each of these approaches has its own advantages and disadvantages.

It has long been known that human T cells isolated from peripheral blood, tumor-draining lymph nodes, or tumor tissue have selective anti-tumor activity, and this provided the rationale for using TILs for cancer immunotherapy more than 20 years ago.[50,51] Early studies demonstrated the feasibility of this approach for treating melanoma and RCC.[50] Although TILs provide an individualized approach to immunotherapy, isolating and expanding TILs ex vivo can be cumbersome and costly. Additionally, to maximize their anti-tumor activity following adoptive transfer, many investigators co-administered high-dose IL-2, which can cause significant toxicity.[49] Results from several studies have shown impressive clinical responses in patients with metastatic melanoma treated with TILs,[52–54] and responses can be enhanced further by lymphodepletion prior to reinfusion of TILs.[53–54] Presumably, agents that drastically reduce lymphocytes also deplete Tregs that can suppress the activity of TILs. Studies are ongoing to identify more efficient and safer methods to culture and administer TILs and to overcome tumor-induced immunosuppression.

Another approach to adoptive T-cell therapy is the use of genetically engineered TCRs. Genes encoding TCRs with specificity for TAAs in the context of common HLA alleles are transduced into $CD4^+$ and $CD8^+$ peripheral blood lymphocytes, thus generating tumor-specific T cells.[51] Any patient whose tumor expresses the correct HLA allele and the target antigen recognized by the TCR could potentially benefit from this therapy. Although initial studies have demonstrated some clinical benefit of

TCRs in patients with metastatic melanoma, the treatment has been associated with significant toxicity to normal tissues that also express the target antigen.[55] Advancing this approach will require the identification of more tumor-specific target antigens.

T cells also can be engineered to express a CAR, which combines the extracellular portion of an antibody with the signaling capabilities of the TCR, thus expanding the spectrum of tumor antigen recognition.[56] Several generations of CARs have been developed. Despite encouraging early clinical results with second-generation CARs in patients with lymphoma, this approach also has been associated with significant toxicity.[57–61] Current studies are aimed at reducing toxicity while maintaining efficacy.

IMMUNE CHECKPOINT INHIBITORS

Numerous inhibitory pathways are built into the immune system to maintain self-tolerance and homeostasis, and these are collectively referred to as immune checkpoints.[62] The primary role of immune checkpoints is to protect tissues from damage when the immune system is responding to pathogens and to maintain tolerance to self-antigens (ie, prevent autoimmunity). This is primarily achieved by down-regulating T-cell activation or effector functions. A growing body of evidence demonstrates that a primary mechanism by which tumors evade the immune system is by engaging immune checkpoints. This has spurred the development of many novel agents that modulate immune checkpoints or other co-stimulatory receptors (Table 2).

The first checkpoint receptor to be successfully targeted as an immunotherapy is CTLA-4.[63] It is expressed on activated T cells, and its primary function is to down-regulate the extent of T-cell activation by countering the co-stimulatory signal delivered by CD28.[64–66] Both CTLA-4 and CD28 share the same ligands, CD80 (also known as B7.1) and CD86 (also known as B7.2); however, CTLA-4 has a higher affinity for these ligands and thus outcompetes CD28 for ligand binding, thereby dampening and limiting the T-cell response.[67–70] The critical role of CTLA-4 in keeping T-cell activation in check is demonstrated by the lethal systemic immune hyperactivation phenotype of *CTLA-4*-knockout mice.[71,72] Ipilimumab, an anti-CTLA-4 monoclonal antibody, was the first immune checkpoint inhibitor to receive FDA approval for the treatment of advanced melanoma.[73] Tremelimumab, another anti-CTLA-4 monoclonal antibody, is in phase II development for mesothelioma and a variety of other solid tumors. These antibodies bind to CTLA-4 and block its immune-suppressive signal. As a result, activated T cells, including those activated by tumor antigens can continue to proliferate, produce cytokines, and exert their cytotoxic effector functions in the tumor microenvironment. Unfortunately, this can sometimes lead to undesired immune-related adverse events caused by an autoimmune reaction to normal tissues.

Targeting PD-1, another immune checkpoint receptor, and its ligands, programmed cell death ligand 1 (PD-L1) and PD-L2, is also emerging as a promising immunotherapeutic modality.[63] Similar to CTLA-4, PD-1 plays a key role in regulating and maintaining the balance between T-cell activation and immune tolerance.[74,75] Unlike CTLA-4, however, PD-1 is broadly expressed and can be found, in addition to T cells, on B cells and NK cells.[76,77] While CTLA-4 primarily regulates T-cell activation in the lymphatic tissues, the main role of PD-1 is to limit T-cell activity in peripheral tissues during a cell-mediated or inflammatory immune response.[74,75,78–82] Tumors can exploit this checkpoint and render TILs, particularly CTLs and NK cells, anergic and unable to kill. The ligand PD-L1 is commonly upregulated on several human solid tumors including melanoma, lung, and ovarian tumors.[83,84] Mice lacking the *Pdl*, *Pdl1*, and *Pdl2* genes have a milder autoimmune phenotype than *CTLA-4*-knockout mice.[80,81,85,86] Several monoclonal antibodies targeting PD-1 (nivolumab [BMS-936558], pidilizumab [CT-011], and lambrolizumab [MK-3475]) and PD-L1 (BMS-936559, MPDL3280A, MSB0010718C, and MEDI4736) are in various stages of clinical development (Table 2). An anti-PD-1 fusion protein, AMP-224, is also in early clinical development. Although no agents targeting PD-1 or its ligands have gained FDA approval yet, clinical studies to date have yielded encouraging results with significant durable response rates in several tumor types, particularly melanoma, RCC, and lung cancer.

Another regulator of the immune response that functions in the tumor microenvironment is the IDO pathway. IDO normally functions to prevent damage from excessive immune activation by breaking down tryptophan, which is required for T-cell activity.[87] Tumor cells often exploit this pathway by overexpressing IDO in the presence of effector cell stimuli. Thus, IDO inhibitors have the potential to alter the tumor microenvironment and boost the T-cell–mediated immune response.

CO-STIMULATORY RECEPTORS

In contrast to the B7/CTLA-4 and PD-L1/PD-1 interactions that suppress T-cell activation, several co-stimulatory receptors are being explored as therapeutic targets. Agents capable of activating co-stimulatory receptors such as OX40 and 4-1BB have entered clinical development recently (Table 2). Preclinical research indicates that these agents may

Table 2. Immune Checkpoint Modulators in Clinical Development

			Clinical Development Status by Tumor Type			
Target		**Drug Name**	**Phase I**	**Phase II**	**Phase III**	**Approved**
Checkpoint Inhibitors	CTLA-4	Ipilimumab (Yervoy, Bristol-Myers Squibb Co, Princeton, NJ))	Pancreatic	Gastric, ovarian	NSCLC, SCLC, prostate	Melanoma
		Tremelimumab	–	Mesothelioma, solid tumors	–	–
	PD-1	MK-3475 (lambrolizumab)	Solid tumors, MM, hematologic	RCC, CRC	NSCLC, melanoma	–
		Nivolumab (BMS-936558)	HCC, CRC, prostate, hematologic	Brain, NHL, esophageal, solid tumors	Melanoma, NSCLC, RCC	–
		Pidilizumab (CT-011)	–	Melanoma, DLBCL-NHL, AML, iNHL	–	–
		MEDI0680 (AMP-514)	Solid tumors	–	–	–
	PD-L1	MEDI4736	–	Melanoma, solid tumors	NSCLC	–
		RG7446 (MPDL3280A)	Melanoma, solid tumors	RCC, bladder	NSCLC	–
		BMS-936559	Solid tumors	–	–	–
		MSB0010718C	Solid tumors	–	–	–
	PD-L2	AMP-224	Solid tumors	–	–	–
	KIR	Lirilumab (BMS-986015)	Solid tumors	AML	–	–
	IDO	Indoximod	Pancreatic, melanoma, brain	Breast, prostate	–	–
Co-stimulatory proteins	OX40	Anti-OX40	–	Prostate	–	–
	4-1BB (CD137)	Urelumab (BMS-663513)	B-cell malignancies, solid tumors	Melanoma	–	–
Other proteins	LAG-3 (CD223)	BMS-986016	Solid tumors, CLL, HL, NHL	–	–	–
	Phosphatidylserine	Bavituximab	Rectal, breast, liver, melanoma	Pancreatic, breast, NSCLC,	–	–

Abbreviations: AML, acute myeloid leukemia; CLL, chronic lymphocytic leukemia; CRC, colorectal cancer; CTLA-4, cytotoxic T-lymphocyte-associated antigen 4; DLBCL, diffuse large B-cell lymphoma; HCC, hepatocellular carcinoma; HL, Hodgkin lymphoma; IDO, indoleamine-2,3-dioxygenase; iNHL, indolent non-Hodgkin lymphoma; KIR, killer cell immunoglobulin—like receptor; LAG-3, lymphocyte-activation gene 3; MM, multiple myeloma; NHL, non-Hodgkin lymphoma; NSCLC, non-small-cell lung cancer; PD-1, programmed cell death 1; PD-L1, programmed cell death ligand 1; PD-L2, programmed cell death ligand 2; RCC, renal cell carcinoma; SCLC, small-cell lung cancer.

Note: This is not a complete list of immune checkpoint modulators in clinical development.

Source: ClinicalTrials.gov.

be most beneficial when given in combination with immune checkpoint inhibitors.[50,88,89] OX40 is a member of the TNF receptor superfamily and is predominantly expressed on $CD4^+$ and $CD8^+$ T cells.[89] Preclinical studies in immunogenic tumor models have demonstrated that an agonist monoclonal antibody to OX40 enhances anti-tumor immunity, and results from a recent phase I trial were encouraging.[90] Agonist monoclonal antibodies against 4-IBB, another member of the TNF receptor superfamily, also have shown promising anti-tumor effects in preclinical studies; however, these agents have not yet entered clinical development.[88] Inducible co-stimulator (ICOS) is another T cell-specific

co-stimulatory molecule that is a member of the CD28/CTLA-4 family.[91,92] T-cell activation results in upregulation of ICOS, and this upregulation is further enhanced upon CTLA-4 blockade.[93,94] A recent study demonstrated that concomitant ICOS stimulation and CTLA-4 inhibition enhanced anti-tumor responses in preclinical mouse models of melanoma and prostate cancer, thus supporting the potential of this combination approach.[95] Likewise, agonists of TLRs regulate the activity of APCs and can have both immune stimulatory or immune inhibitory effects depending on whether they induce expression of co-stimulatory molecules (eg, CD80, CD86, and CD40) and proinflammatory cytokines (eg, TNF-α and IL-12) or instead induce expression of co-inhibitory molecules (eg, IL-10 and PD-L1).[96] Studies have demonstrated that concomitant blockade of IL-10 or PD-L1 can augment the anti-tumor effects of TLR agonists.

CONCLUSIONS

Cancer immunotherapy has been studied and tested for several decades, but only recently have immune-based therapies been shown to provide an OS benefit in patients with advanced cancer. For many years, success was limited to a select few patients. As our understanding of the complex interactions between tumors and the immune system has advanced, a range of new therapeutic strategies has been developed. These new agents exploit a wide array of mechanisms to enhance the anti-tumor immune response and are beginning to have a clinical impact on the treatment of many different tumor types. Clinical results of approved and emerging immunotherapies will be discussed further in the articles by Weber and Rini within this supplement.

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