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REVIEW

TIM-3, a promising target for cancer immunotherapy

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Abstract: Patients with malignant tumor treated with immunotherapy have received significant clinical benefits over the years. Immune checkpoint blocking agents, such as anti-cytotoxic T-lymphocyte-associated protein-4 (anti-CTLA-4) and anti-programmed cell death protein-1 (anti-PD-1) monoclonal antibodies, have produced impressive clinical results in different types of cancer. T-cell immunoglobulin and mucin domain-3 (TIM-3), another immune checkpoint, could inhibit cancer immunity. Recent studies have highlighted that TIM-3 has an important role to play in T-cell exhaustion and correlates with the outcome of anti-PD-1 therapy. Targeting TIM-3 might be a promising approach for cancer immunotherapy. Here, we review the role of TIM-3 in cancer and clinical trials with TIM-3 inhibitors.

Keywords: immune checkpoint, clinical trial, cancer immunotherapy, T-cell immunoglobulin and mucin domain-3 (TIM-3)

Background

In recent years, cancer immunotherapy, such as programmed death receptor 1 (PD-1) and programmed death-ligand 1 (PD-L1) monoclonal antibodies, has shown promising therapeutic outcomes in cancer.[1–5] T-cell immunoglobulin mucin-3 (TIM-3) is another important cancer immune checkpoint.[6] Patients treated with anti-PD-1 or anti-PD-L1 monoclonal antibodies will face the resistance problems. Koyama et al[6] reported TIM-3 expression was increased when patients faced the anti-PD-1 adaptive resistance.

Introduction to TIM-3

TIM-3, also known as HAVCR2, belongs to the *TIM* gene family. In humans, the TIM family includes TIM-1, TIM-3, and TIM-4 and is located on chromosome 5q33.2. In mice, the TIM family includes TIM-1 to TIM-8 and is located on chromosome 11B1.1.[7]

TIM-3, as a negative regulatory immune checkpoint, is detected in different types of immune cells, including T cells, regulatory T cells (Tregs), dendritic cells (DCs), B cells, macrophages, nature killer (NK) cells, and mast cells.[7–9] TIM-3 is a type I membrane protein and consists of 281 amino acids. It comprises an extracellular domain, a single transmembrane domain, and a C-terminal cytoplasmic tail.[9–13]

TIM-3 has four ligands, including galectin-9 (Gal-9), carcinoembryonic antigen cell adhesion molecule 1 (CEACAM-1), high-mobility group protein B1 (HMGB1), and phosphatidylserine (PS).[14] Gal-9 was the first to be identified. It is a carbohydrate binding protein, specifically recognizing the structure of N-linked sugar chains in the TIM-3 immunoglobulin variable (IgV) domain.[15] TIM-3/Gal-9 can inhibit cancer immunity by negatively regulating T-cell immunity. The connection of the TIM-3 IgV

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domain with Gal-9 can terminate T helper 1 (Th1) immune responses.[10]

TIM-3 could induce immunological tolerance.[10,16] Its molecules are related to asthma, food allergy, and autoimmune disease, such as multiple sclerosis and rheumatoid arthritis.[7,16] TIM-3 could also inhibit the immune responses of T cells and was associated with immune exhaustion, which induced chronic viral infection.[12,13,15]

TIM-3 and cancer immunity

TIM-3 inhibited antitumor immunity by mediating T-cell exhaustion.[15] TIM-3+ $CD8^+$ T cells exhibit impaired Stat5 and p38 signaling pathway. Blocking the TIM-3 pathway enhanced cancer immunity and increased the production of interferon-gamma (IFN-γ) in T cells.[17] In in vitro and in vivo models, the expression of $CD8^+$ TIM-3+ T cells was correlated with PD-1 expression. TIM-3 was constitutively expressed on innate immune cells and could suppress innate antitumor immunity. TIM-3 inhibited the proliferation and effector of cytokine production, such as interleukin-2 (IL-2).[18–20] PD-1 and TIM-3 positive $CD8^+$ T cells produced less IFN-γ than TIM-3 negative $CD8^+$ T cells.[21] Anti-TIM-3 antibodies could also increase IFN-γ of peripheral NK cells.[22] Mast cells expressing TIM-3 could be activated through an ITAM-containing receptor for IgE (FcepsilonRI), using signaling pathways analogous to those in T cells. TIM-3 acts at a receptor-proximal point to enhance Lyn kinase-dependent signaling pathways that modulate both immediate-phase degranulation and late-phase cytokine production downstream of FcepsilonRI ligation.[9] TIM-3 could be detected in non-small cell lung cancer (NSCLC),[22,23] hepatocellular carcinoma (HCC),[24] colorectal cancer,[24–28] cervical cancer,[29] ovarian cancer,[24,30] head and neck cancer,[31] and so on.

In myelogenous leukemia (AML), upregulated TIM-3 during AML could reduce cytokine production. Co-expression of PD-1 and TIM-3 was correlated with AML progression.[18] In follicular B-cell non-Hodgkin lymphoma, TIM-3 was expressed on nearly 35% of lymph node $CD4^+$ and $CD8^+$ T cells and could mediate T-cells exhaustion.[32] In glioma patients, TIM-3 was correlated with cancer immune escape and might be a potent target.[33] In gastric cancer, TIM-3 could promote disease progression,[34] and Gal-9 and TIM-3 expressed on tumor cells might be a potential, independent prognostic factor. Decreased Gal-9 and increased TIM-3 were associated with a poor prognosis in gastric cancer.[35] PD-1+ and TIM-3+ $CD8^+$ T cells could impair the functioning of $CD8^+$ T cells in gastric cancer.[21,36] In colorectal cancer, upregulation of TIM-3 could restrict T-cell responses and might participate in tumorigenesis. The expression of TIM-3 might be an independent prognostic factor for colorectal cancer.[27] TIM-3 was correlated with the progression of colorectal cancer and could be a potential therapeutic target for the disease.[25] PD-1 and TIM-3 could impair surgery colorectal cancer patients' cell-mediated immunity.[28] In NSCLC patients, TIM-3 was expressed on about 30% of $CD8^+$ tumor-infiltrating lymphocytes (TILs) and 60% of $CD4^+$ FoxP3+ TILs. TIM-3+ FoxP3+ Tregs were correlated with the lung cancer stages.[37] TIM-3 expression in NK cells was related to disease progression of lung cancer.[38] In prostate cancer, TIM-3 could affect disease development and progression.[39,40] In renal cell carcinoma (RCC), TIM-3 expressed on cancer cells and in myeloid cells could inhibit cancer immunity.[41] In ovarian cancer, TIM-3 could negatively regulate various T-cell subsets. TIM-3 expression on $CD4^+$ T cells could serve to predict the outcome of anticancer therapies.[30] In cervical cancer, the expression of TIM-3 in tumor cells might be a potential prognostic factor and could promote metastases.[29]

Targeting TIM-3 in cancer

TIM-3 could be a promising target in cancer because of its expression on a variety of T cells.[16] TIM-3 was also expressed on myeloid cells, such as DCs, macrophages, and monocytes. TIM-3 has an important role in innate immune cell-mediated antitumor immune responses.[16,42]

An increasing number of preclinical studies have reported that TIM-3 could improve the outcomes of cancer immunotherapy (Table 1).

TIM-3 inhibitors have shown similar efficacy as that of PD-1 inhibitors in preclinical research.[44] It was reported that PD-1 antibodies may lead to an increase in TIM-3 expression in in vivo models of lung cancer, which showed TIM-3 might be a marker of PD-1 blocking antibody resistance.[6] PD-1, TIM-3, and LAG-3 were upregulated on tumor-associated antigen-specific T cells in HCC tissues. PD-1, TIM-3, or LAG-3 inhibitors could enhance T cells' response to tumor antigens, and had a synergistic function.[52] TIM-3+ PD-1+ $CD8^+$ TILs inhibited the production of cytokines, such as IFN-γ, tumor necrosis factor-alpha (TNF-α), and IL-2.[51] The combined use of TIM-3 blockade with PD-1 blockade could be more effective than blockade of either the TIM-3 or PD-1 alone.[6,17–19,43,44,48,49,51,53]

Currently, many clinical trials are focusing on TIM-3 as a new approach to the treatment of cancer (Table 2).

Cancer immunotherapy has shown promising therapeutic outcomes. T-cell checkpoint inhibitor is one of the most promising new therapeutic approaches in cancer. TIM-3

Table 1 TIM-3 and cancer

Year	Diseases	Conclusions	References
2010	Solid tumors	Combined TIM-3 with PD-1 inhibitor could prevent tumor progression.	19
2010	Melanoma	TIM-3/TIM-3L inhibitor combined with PD-1/PD-L1 inhibitor could reverse T-cell exhaustion and/or dysfunction in advanced melanoma.	43
2011	Cancer	Anti-TIM-3 molecular antibody suppressed tumors by promoting T-cell IFN-γ-mediated antitumor immunity.	44
2011	AML	Combined PD-1/PD-L1 with TIM-3/Gal-9 blockade could prevent $CD8^+$ T-cell exhaustion in advanced AML.	18
2013	AML	In xenograft models, anti-TIM-3 IgG2a antibody could improve cytotoxic activities and eradicate AML leukemic stem cells.	45
2013	Melanoma	Combined anti-TIM-3 with anti-TIM-4 molecule antibodies could increase the antitumor responses in vivo.	46
2013	Ovarian cancer	Combined anti-TIM-3 and CD137 molecule antibodies significantly inhibited tumor progression.	47
2014	Melanoma	PD-1 combined with TIM-3 blockades could stimulate potential antitumor T-cell responses in melanoma.	48
2015	Gastric cancer	Combined treatments of TIM-3 and CD137, TIM-3 and PD-1, and TIM-3 and CEACAM1 could enhance immune cell response in progression stage cancer. And anti-TIM-3 and anti-TIM-4 molecule antibodies could increase cancer vaccine's efficacy.	49
2015	RCC	TIM-3 expressed on myeloid cells played a critical role in augmenting tumorigenic activities of TIM-3-negative RCC cells. Anti-TIM-3 monoclonal antibody suppressed the cancer cells.	41
2015	Colon cancer	Gal-9/TIM-3 blockade could inhibit the tumor progression in vivo. The blockade increased therapeutic efficacy of cyclophosphamide.	50
2015	Colon cancer	TIM-3 was correlated with colon cancer immune escape.	26
2015	Lung adenocarcinoma	TIM-3 could express on NK cells and was a potential new immune therapy target.	22
2015	Colorectal carcinoma	Higher expression of TIM-3 indicated restriction of T-cell responses.	27
2015	Gastric cancer	TIM-3 expression was correlated with the stages of gastric cancer and was regulated by T-bet.	36
2016	RCC	Blocking the TIM-3 pathway reversed cell proliferation and increased IFN-γ production in varied types of T cell.	17
2016	Colorectal carcinoma	TIM-3/TIM-3L and PD-1/PD-L1 blockade reversed T-cell dysfunction and exhaustion in colorectal cancer.	51
2016	Glioma	Gal-9/TIM-3 pathway was important in immune evasion and could be a potential target in glioma.	33
2017	AML	TIM-3/Gal-9 was a reliable target for AML immune therapy.	20
2017	HCC	Antibodies against PD-L1, TIM-3, or LAG-3 restored responses of HCC-derived T cells to tumor antigens.	52
2017	Gastric cancer	Dual blockade of TIM-3 and PD-1 could improve antitumor function of cancer $CD8^+$ T cells.	53
2017	Colorectal cancer	TIM-3 was correlated with the progression of colorectal cancer and could be a potential therapeutic target.	25
2017	Prostate cancer	TIM-3 inhibited the immune response in prostate cancer and could be a potential therapeutic target.	40

Abbreviations: TIM-3, T cell immunoglobulin mucin-3; TIM-3L, T cell immunoglobulin mucin-ligand 3; PD-1, programmed cell death protein-1; PD-L1, programmed cell death protein-ligand 1; IFN-γ, interferon-γ; Gal-9, galectin-9; AML, acute myeloid leukemia; RCC, renal cell carcinoma; NK, nature killer; HCC, Hepatocellular carcinoma; LAG-3, lymphocyte-activation gene-3.

inhibits antitumor immunity. The roles of TIM-3 in cancer immunity need to be further investigated. New treatment targeting TIM-3 could soon provide a breakthrough in cancer treatment and improve patient outcomes.

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Table 2 Clinical trials of TIM-3 inhibitors

Year	Drug	Phase	Company	Type	Objective	ClinicalTrial.gov identifier
2015	MBG453	I	Novartis Pharmaceuticals (Basel, Switzerland)	Anti-TIM-3	MBG453 given alone or combined with PDR001 in adult patients with advanced malignancies	NCT02608268
2016	TSR-022	I	Tesaro, Inc. (Waltham, MA, USA)	Anti-TIM-3	Dose escalation and cohort expansion study of TSR-022 in advanced solid tumors	NCT02817633
2017	LY3321367	I	Eli Lilly and Company (Indianapolis, IN, USA)	Anti-TIM-3	LY3321367 alone or combined with an anti-PD-L1 antibody in advanced relapsed/refractory solid tumors	NCT03099109
2017	MBG453	I	Novartis Pharmaceuticals	Anti-TIM-3	PDR001 and/or MBG453 in combination with decitabine in AML or high-risk MDS	NCT03066648

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Disclosure

The authors report no conflicts of interest in this work.

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