TIM-3: a tumor-associated antigen beyond checkpoint inhibition?

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Summary

Immune checkpoint inhibitors are one of the most remarkable immunomodulatory therapies of current times. Sabatolimab is a high-affinity, humanzed anti-TIM-3 monoclonal antibody currently in development for patients with myeloproliferative disorders, including acute myeloid leukemia and myelodysplastic syndromes. By targeting TIM-3, a receptor expressed on various immune effector cells as well as myeloid cells, multiple mechanisms of action that are distinct from canonical immune checkpoint inhibitors are in play – (i) blockade of TIM-3 and its ligands PtdSer/galectin-9, (ii) modulation of leukemic cell self-renewal as well as (iii) antibody-dependent phagocytosis of TIM-3–expressing leukemic cells. Novel immunotherapies such as sabatolimab which enhance the antitumor immune response on converging fronts represent the promise of a continuously replenished armory for the treatment of cancer.

Keywords: TIM-3, tumor-associated, antigen, checkpoint, inhibition

Immunotherapy harnesses the host’s adaptive and innate immune response to effectuate the direct targeting and elimination of diseased cells via specific, mechanism-based agents. This approach has transformed cancer treatment over the last two decades as reflected by the steadily increasing number of approved antibodies clinically available, which either enhance T-cell effector function or directly neutralize diseased cells.

Antibodies are highly selective proteins are produced by B cells representing the human adaptive humoral immune system which serves to protect the host against various sources of threat, including foreign pathogens as well as our own dysregulated cancerous cells. The ground-breaking, Nobel prize-winning work of Jerne, Kohler and Milstein on hybridoma technology [1, 2] propelled antibodies into easily accessible and highly dynamic key players that constantly redefine their diagnostic and/or therapeutic application. Furthermore, antibodies selectively target uniquely expressed cell surface receptors, a feature that has been exploited for development of cancer therapeutics targeting surface receptors on tumor cells and allowing direct selective targeting [3]. FDA approved antibody therapeutics for cancer include targeting e.g. CD19, CD20, CD22, CD30, CD33, CD38, CD52, CD79b, CD138, CRR4, EGFR, GD2, HER2, Nectin-4, PDGFR, RANKL, SLAMF7, TF, TROP-2, VEGFR via naked monoclonal antibodies or antibody drug conjugates. Furthermore, the versatility of antibody therapies has been described as an efficient approach for the treatment of infectious diseases [4, 5] as recently demonstrated for the current COVID-19 pandemic via the development of approved SARS-CoV-2 neutralizing antibodies [6–8].

Targeting of co-inhibitory receptors cytotoxic T lymphocyte associated protein 4 (CTLA-4) and programmed death ligand 1 (PD-1) or its ligand PD-L1 has transformed oncology and led to long-term durable remissions across many cancer types [9, 10]. Activation of effector T cells is driven by stimulation of different sets of receptors: (i) activation is mainly driven by stimulation of the T-cell receptor by MHC-presented peptides together with co-stimulatory receptors engaging with their corresponding ligands on antigen presenting cells, (ii) inhibition is controlled by interaction of co-inhibitory receptors including among others PD-1, or CTLA-4 with their natural ligands. Consequently, blockade of such immune checkpoint receptors by inhibitors has emerged as a prominent treatment option for nearly half of all patients with metastatic cancers. The co-inhibitory receptor LAG-3 is the most recent checkpoint blockade pathway to receive FDA approval (PMID: 35265944, PMID: 34986285). However, with the durable responses of these therapies, the long-term implications of acute clinical toxicities associated with these agents are of growing concern [11, 12].

In this issue of Immunotherapy Advances, Schwartz et al. exploits the unique dual role of T-cell immunoglobulin and mucin containing protein 3 (TIM-3) – originally described as a cell surface marker specifically expressed by CD4+ T helper 1 and CD8+ cytotoxic T cells [13] and recently confirmed as an immune checkpoint receptor [14]. Furthermore, the
specific role of TIM-3 in regulating immune responses at sites of tissue inflammation is a feature distinct to co-inhibitory receptors CTLA-4 ad PD-1 which are primarily involved in maintaining self-tolerance [15]. The upregulation of TIM-3 on exhausted T cells in cancer has provided a rationale for the development of TIM-3 blocking antibodies for immune checkpoint blockade. Sabatolimab an anti-TIM-3 antibody developed by Novartis is currently tested as single agent or in combination with other therapies in early phase trials. Here, Schwartz et al. present, a promising mechanistic in vitro analysis of multiple mechanisms of action of sabatolimab, highlighting roles for TIM-3 as a stem cell antigen in myeloid cell leukemias and as a tumor-associated antigen upregulated in myeloproliferative syndromes (MDS) and myeloproliferative neoplasms (MPN) [16]. Most surprisingly, TIM-3 does not appear to inhibit T cell effector functions. Their main conclusions are as follows:

1) Sabatolimab blocks binding of TIM-3 to its ligands Galectin-9 and PtdSer: A TIM-3/galectin-9 interaction might contribute to disease progression in MDS, leukemic transformation and self-renewal of leukemic stem cells. In a Luminex assay, binding of phycoerythrin-labeled galectin 9 to TIM-3-Fc decorated bead was blocked by sabatolimab in a dose-dependent manner. Crystal structure data also reveal that the PtdSer binding site is occluded by the antibody.

Enhanced T-cell killing requires TIM-3 expression on the tumor cells, not on the T cells: Using TIM-3-positive HNT-34 acute myeloid leukemia (AML) cells co-cultivated with CD3-activated PBMCs from different healthy donors, sabatolimab-driven blockade enhanced immune-mediated killing of the leukemic cells in a donor-dependent manner. Interestingly, the Sabatolimab enhancement was observed with TIM-3 overexpressing AML cells, but not with parental AML cells, highlighting an Fc-driven T-cell-mediated killing.

2) TIM-3 is functional on human dendritic cells: TLR activation of isolated human DCs showed that sabatolimab-blockade of TIM-3 enhanced the secretion of IL-6, IL-12, and TNFα, indicating that TIM-3 is a negative regulator of DC activation.

3) Sabatolimab facilitates opsonization of TIM-3 expressing tumor cells: Antibody-dependent cellular phagocytosis (ADCP) was confirmed after addition of sabatolimab to TIM-3 overexpressing Raji cells co-incubated with THP-1-derived phagocytic cells. Importantly, sabatolimab treatment of phagocytes did not abrogate ADCP activity, and TIM-3-negative parental Raji cells did not induce any ADCP. Together these studies show that TIM-3 expression on tumor cells allows for sabatolimab binding and enhanced phagocytosis by macrophages. Similar results were observed using monocyte-derived macrophages co-cultured with TIM-3 expressing HNT-34 or SKM-1 cell lines.

This study provides the first biophysical characterization of sabatolimab, and an insightful glimpse into the mechanistic role of TIM-3. Although originally classified as an inhibitory receptor on T cells, Schwartz et al. clearly show that sabatolimab does not enhance T-cell effector functions unless the tumor cells express TIM-3. Dendritic cell expression of TIM-3 appears to be an important negative regulator of cytokine production, at least in the context of TLR stimulation. The biggest role for TIM-3 in AML may be as a tumor antigen where sabatolimab blockade of TIM-3 binding to its ligands and enhanced opsonization combine to eradicate leukemic cell growth. Whether these diverse activities will indeed result in improved clinical outcomes remains to be demonstrated, but the outlook is hopeful. The advent of immunotherapies has revolutionized cancer treatment, but with much room left for improvement, the discovery of novel agents like sabatolimab continue to supplement the current portfolio of therapeutic options.

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Conflicts of interest
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Not applicable.

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