



Overcoming Resistance to Immune Checkpoint Blockade: Mechanistic Insights and Emerging Therapeutic Directions

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ABSTRACT

Immune checkpoint inhibitors have revolutionized cancer therapy by delivering long-lasting responses in a subset of patients across many cancer types. Yet, their effectiveness is often limited by high rates of primary and acquired resistance. This resistance is driven by complex interactions among tumor-intrinsic alterations, immunosuppressive factors within the tumor microenvironment, and host-related determinants. This review critically examines the biological mechanisms underlying resistance to immune checkpoint inhibitors, including defects in antigen presentation, dysregulated interferon signaling, activation of oncogenic pathways, compensatory upregulation of alternative immune checkpoints, and microbiome-associated immune modulation. In addition to defining these challenges, this review also highlights emerging opportunities for overcoming these obstacles. Emerging opportunities to circumvent resistance include biomarker-guided patient stratification, rational combination therapies that engage complementary immune pathways, modulation of the tumor microenvironment, and integration of multi-omics approaches to identify predictive resistance signatures. The central conclusion of this review is that effective clinical translation will necessitate a paradigm shift from discrete pathway inhibition toward integrated precision immuno-oncology strategies that combine molecular profiling, immune-contexture analysis, and mechanism-based combination therapies. Such integrated approaches may improve patient selection, overcome resistance, and expand the proportion of patients who achieve durable responses to immune checkpoint blockade.

Keywords: Cancer, Immune checkpoints, Immunotherapy, Monoclonal antibodies

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INTRODUCTION

Over the course of the last decade, researchers have achieved by researchers in the domain of cancer immunotherapy by employing complementary strategies that entail the blockade of immune system inhibitors (1). The study examines therapeutic options that aim to enhance the immune system's capacity to fight against cancer by selectively targeting different pathways that impede immune activity. These pathways encompass several mechanisms, such as the modulation of immune-regulating cytokines, tumor vaccines, allogeneic stem cell transplantation, and adoptive cell transfer (2, 3). The utilization of immune checkpoint blockade therapy has emerged as a groundbreaking approach to eliminating tumor cells by alleviating constraints on the immune response to malignancies (4). Monoclonal antibodies have been instrumental in revealing the profound influence of key immune checkpoints—such as programmed cell death protein-1 (PD-1), cytotoxic T-lymphocyte antigen 4 (CTLA-4), lymphocyte activation gene 3 (LAG-3), and T cell immunoglobulin and mucin domain-containing protein 3 (TIM-3)—on the trajectory of various cancers (4, 5).

The use of immune checkpoint inhibitors that selectively target CTLA-4 and PD-1, as well as the PD-1 ligand PD-L1, particularly Ipilimumab, Pembrolizumab, Nivolumab, and Atezolizumab, has been employed for the treatment of malignant tumors, resulting in significant enhancements in patient survival (6, 7). This therapeutic strategy is now a standard approach for a range of medical conditions, such as non-small cell lung cancer (8), colon cancer (9), esophageal cancer (10), melanoma (11), renal cell carcinoma (12), bladder carcinoma (13), and breast cancer (14). Nevertheless, the development of resistance to these inhibitors poses a significant barrier to the effective implementation of cancer immunotherapy. Hence, it is imperative to elucidate the mechanisms that underpin drug resistance.

Numerous reviews have described the biological basis of resistance to immune checkpoint blockade (ICB), yet the rapidly expanding literature has left us with a fragmented picture of how tumor-intrinsic, tumor-extrinsic, and host-related factors interact to shape therapeutic response. Most prior reviews have focused on isolated mechanisms—specific checkpoint pathways, signaling alterations, or components of the tumor microenvironment—without building an integrated framework that connects these mechanisms to translational opportunities in clinical oncology. A fresh synthesis is now warranted: recent advances in multi-omics profiling, spatial immunology, microbiome research, and biomarker-driven clinical trial design have substantially expanded our understanding of ICB resistance, but a critical gap remains in translating these findings into a clinically relevant model that can guide patient stratification, biomarker development, and rational combination therapy. This review therefore examines ICB resistance mechanisms across three interacting categories—tumor-intrinsic, tumor-extrinsic (microenvironmental), and host-related (systemic and germline) factors—with emphasis on their crosstalk and implications for future clinical trials and precision immunotherapy. By integrating mechanistic insights with translational considerations, we aim to provide a more structured and clinically actionable perspective than previous descriptive summaries.

IMMUNE CHECKPOINT BLOCKADE IN CANCER THERAPY

Programmed death-1 (PD-1)

The co-inhibitory receptor PD-1, alternatively referred to as CD279, exhibits broad expression on T and B cells, as well as limited expression on natural killer (NK) cells. PD-1, which is significantly upregulated on activated T cells, is widely acknowledged to play a crucial role in the

formation and preservation of immunological tolerance (15). PD-L1 and PD-L2 are of the two ligands capable of interacting with PD-1. The ligands exhibit low expression in healthy tissue but show significantly elevated expression in specific cancer types. It has been demonstrated that PD-L1 expression is increased in several cancer types, including melanoma, non-small-cell lung cancer (NSCLC), breast cancer, and squamous cell carcinoma of the head and neck (16).

Prior studies have contributed to the emergence of novel approaches that specifically target the inhibition of PD-1 or PD-L1 function. Hence, it has been demonstrated that these methodologies can efficiently reinstate the functionality of T-cells (17). Monoclonal antibodies that specifically bind to PD-1 and its associated ligand, PD-L1, has been employed in the therapeutic intervention of various cancers. Huang et al. (2018) conducted a study that revealed the efficacy of neoadjuvant anti-PD-1 therapy in the treatment of patients diagnosed with resectable stage III/IV melanoma, a condition characterized by a heightened likelihood of recurrence (18). Garon et al. conducted a phase I clinical trial to assess the safety and efficacy of pembrolizumab, an anti-PD-1 monoclonal antibody, in patients with advanced NSCLC. They observed significant anti-tumor activity with manageable adverse effects (19). Zhang et al. employed single-cell RNA sequencing (scRNA-seq) to analyze 3110 peripheral T cells from patients with NSCLC. The study examined the characteristics of tumor-associated CD4⁺ T-cell clones compared with tumor-associated CD8⁺ T-cell clones, both before and during PD-1 inhibition. They showed that tumor-associated CD4⁺ T-cell exhibited greater cytotoxic activity than CD8⁺ T-cell clones (20).

Findings from a longitudinal trial spanning three years on the long-term outcomes of advanced renal cell carcinoma indicate that nivolumab demonstrates superior efficacy and safety compared with everolimus (21). Two phase III clinical trials, CheckMate 141 and

KEYNOTE 040 evaluated overall survival (OS) in patients with recurrent or metastatic head and neck squamous cell carcinoma (HNSCC) (22, 23). Nivolumab significantly improved OS in this population in CheckMate 141 (NCT02105636), while pembrolizumab showed a survival benefit compared with standard therapy in KEYNOTE-040 (NCT02252042). However, these trials also revealed substantial heterogeneity in treatment response, indicating that only a subset of patients derives durable clinical benefit from PD-1 inhibition. This variability supports the involvement of resistance mechanisms such as impaired antigen presentation, defective interferon signaling, and immunosuppressive features of the tumor microenvironment (24). Collectively, the evidence reviewed in this section demonstrates that PD-1/PD-L1 blockade has achieved meaningful clinical success in several malignancies, including melanoma, NSCLC, renal cell carcinoma, and recurrent or metastatic HNSCC, where clinical trials have reported improved survival and manageable safety profiles. However, comparison across studies also reveals substantial heterogeneity in treatment response. While some patients experience durable benefit, others show limited or no response despite receiving the same therapeutic approach. This variability indicates that the efficacy of PD-1 blockade depends on multiple interacting biological factors, including tumor immunogenicity, the extent of T-cell infiltration, antigen presentation capacity, and the immunosuppressive characteristics of the tumor microenvironment. PD-L1 expression, although widely used in patient selection, does not consistently predict treatment outcome across all tumor types. Moreover, the differential clinical benefit observed among malignancies highlights tumor types in which translation has been successful, such as melanoma and NSCLC, as well as those in which it remains less effective, including tumors with poor immune infiltration. Therefore, current evidence suggests that future improvement in PD-1/

PD-L1-targeted therapy will depend on refined patient stratification and rational combination strategies aimed at overcoming complementary resistance mechanisms.

Cytotoxic T-lymphocyte antigen 4 (CTLA-4)

CTLA-4, also known as CD152, is a co-inhibitory receptor expressed on T-cells. Originally identified as a secondary receptor for the B7 costimulatory protein, subsequent studies have established its inhibitory role in T-cell activation (25, 26). Following T-cell receptor (TCR) engagement and stimulation, CTLA-4 is rapidly upregulated on both CD4⁺ helper T cells and CD8⁺ effector T cells, with more pronounced expression on CD4⁺ T cells (27). Its ligands, CD80 (B7-1) and CD86 (B7-2), also known as B7-1 and B7-2, are also recognized by CD28, a costimulatory receptor structurally related to CTLA-4. CTLA-4 exhibits higher affinity and avidity for these ligands than CD28 enabling it to outcompete CD28 and function as a negative regulator of costimulation (28, 29). Collectively, the CD28/CTLA-4 axis serves as a critical checkpoint controlling T cell activation.

In mouse models, anti-CTLA-4 antibodies were the first agents to achieve tumor eradication, including of preexisting tumors, and induced protective immunity against subsequent tumor challenge (30). In clinical development, two anti-CTLA-4 antibodies—ipilimumab and tremelimumab—were evaluated across a broad range of human tumor types. Therapeutic efficacy has been reported in melanoma (31, 32), non-small cell lung cancer (33), mesothelioma (34), prostate cancer (35), breast cancer (36), and urothelial cancer (37). These trials also revealed that approximately 60-65% of patients experience immune-related adverse events (irAEs) affecting the skin, gastrointestinal tract, liver, and endocrine organs, even when therapeutic benefit is observed. The phase III CheckMate 067 trial (NCT01844505) evaluated nivolumab plus ipilimumab versus either agent alone in 945 patients with previously untreated stage

III or IV melanoma. At 6.5 years, median overall survival (OS) was 72.1 months with combination therapy, 36.9 months with nivolumab alone, and 19.9 months with ipilimumab alone (39, 40). However, the enhanced efficacy of combination therapy was accompanied by higher rates of irAEs and treatment discontinuation, highlighting the clinical challenge of balancing therapeutic benefit with treatment-related toxicity.

Collectively, the evidence reviewed in this section indicates that CTLA-4 blockade has played a pivotal role in establishing immune checkpoint blockade as an effective therapeutic strategy, particularly in metastatic melanoma, where trials have demonstrated meaningful survival benefits. However, comparison across preclinical and clinical findings reveals a more complex therapeutic profile than initially suggested by experimental models. Preclinical studies showed robust antitumor activity following CTLA-4 inhibition, whereas clinical trials across different malignancies have demonstrated variable efficacy and a considerable incidence of irAEs. The evidence further suggests that the benefit of CTLA-4-targeted therapy depends strongly on tumor immunogenicity and the presence of a pre-existing antitumor immune response. Combination therapy with CTLA-4 and PD-1 inhibitors has shown enhanced efficacy in selected settings, such as advanced melanoma, but these improvements have frequently been accompanied by increased treatment-related toxicity and higher rates of treatment discontinuation. These observations underscore the need to balance therapeutic efficacy and safety against immune-related toxicity in the clinical application of CTLA-4 blockade. Therefore, current evidence suggests that future optimization of CTLA-4-targeted immunotherapy should focus on improved patient selection and rational combination strategies aimed at maximizing antitumor efficacy while minimizing immune-related toxicity.

T cell immunoglobulin and mucin domain-

containing protein 3 (TIM-3)

TIM-3 was initially identified as a surface molecule selectively expressed on interferon (IFN)-secreting T cells. It is also found on regulatory T cells (Tregs) and innate immune cells, including dendritic cells (DCs), natural killer (NK) cells, and monocytes (41). It is also found on regulatory T cells (Tregs) and innate immune cells, including dendritic cells (DCs), natural killer (NK) cells, and monocytes (41). TIM-3 gene is regulated by both antigenic stimulation and proinflammatory cytokines (42). In animal models, administration of anti-TIM-3 antibodies exacerbated autoimmune encephalomyelitis (43). Recent studies link TIM-3 overexpression to the onset of T-cell dysfunction and exhaustion (44). TIM-3, which suppresses T-cell activity, is a significant factor in chronic viral infections caused by human immunodeficiency virus 1 (HIV-1), hepatitis B virus (HBV), and hepatitis C virus (HCV) (45, 46). TIM-3 is expressed on CD8⁺ tumor-infiltrating leukocytes (TILs), where its expression correlates strongly with PD-1 within the tumor microenvironment. Moreover, increased TIM-3 levels on CD8⁺ T cells correlate with worse prognosis in certain cancer subtypes. Co-blockade of TIM-3 and PD-1 has been shown to augment the efficacy of cancer immunotherapy relative to PD-1 blockade alone (47, 48).

A current study examined the effects of a newly developed Tim-3 antibody on peripheral blood mononuclear cells (PBMCs), T cells, and monocytes/macrophages with the primary objective of evaluating its therapeutic potential TIM-3-associated immunological disorders. The prevailing strategy for assessing therapeutic efficacy involves combining anti-TIM-3 monoclonal antibodies with antibodies targeting LAG-3 and PD-1. Preclinical cancer models have demonstrated a synergistic effect when PD-1 and TIM-3 are simultaneously inhibited, and in murine tumor models, dual blockade of TIM-3 and PD-1 restored T-cell function and enhanced antitumor activity compared with blockade of either pathway alone (50).

Sabatolimab (MBG453) is a high-affinity, humanized anti-TIM-3 monoclonal antibody. Preclinical studies have established that TIM-3 is associated with T-cell exhaustion and may contribute to adaptive resistance following PD-1 blockade. Early-phase clinical evaluation of sabatolimab in combination with spartalizumab (anti-PD-1; NCT02608268) demonstrated acceptable tolerability and preliminary antitumor activity, although definitive evidence from randomized clinical trials remains limited (51). Moreover, anti-TIM-3 antibodies such as sabatolimab, in combination with other checkpoint inhibitors, has exhibited effectiveness as a prospective approach for immunotherapy in represent a promising immunotherapeutic approach in myeloid malignancies, including myelodysplastic syndromes and acute myeloid (52).

Collectively, the evidence reviewed in this section indicates that TIM-3 is a promising immune checkpoint target because of its association with T-cell exhaustion and its frequent co-expression with PD-1 in the tumor microenvironment. Preclinical studies consistently support simultaneous inhibition of TIM-3 and PD-1, demonstrating enhanced restoration of T-cell function and improved antitumor activity compared with blockade of either pathway alone. However, comparison of these experimental findings with available clinical observations reveals that the translation of TIM-3-targeted therapy into meaningful clinical benefit remains limited. Early-phase clinical studies have generally shown acceptable tolerability, but TIM-3 inhibition as monotherapy has produced modest or nonsignificant antitumor effects, whereas combination approaches have shown more encouraging activity in selected patient subsets. This discrepancy suggests that TIM-3 may contribute more prominently to adaptive resistance mechanisms, particularly following PD-1 blockade, than to primary immune suppression across all tumor settings. Furthermore, the broad expression of TIM-3 across multiple immune cell populations

indicates that its biological effects are context dependent and may vary by tumor type and immune microenvironment. Therefore, current evidence suggests that future development of TIM-3-targeted therapy should prioritize biomarker-guided patient selection and rational combination strategies rather than reliance on TIM-3 inhibition as a standalone approach.

Lymphocyte activation gene 3 (LAG-3)

LAG-3 (CD223) was cloned in 1990 as a transmembrane protein upregulated on activated T lymphocytes and NK cells (53). LAG-3 (CD223) was cloned in 1990 as a transmembrane protein upregulated on activated T and NK cells. It is a recently identified immune checkpoint that shares structural homology with CD4 and binds major histocompatibility complex (MHC) class II with higher affinity than CD4 (54). LSECtin, a DC-SIGN family member, has also been proposed as a LAG-3 ligand (55). The primary recognized function of LAG-3 is inhibition of T cell responses, positioning it as a promising target for immunomodulation. Both natural Tregs (nTreg) and induced FoxP3⁺ Tregs (iTreg) express LAG-3, and LAG-3 blockade impairs their suppressive function (56). Prior studies support a role for LAG-3 in modulating T cell homeostasis, a process controlled in part by Tregs (57, 58).

Prior research has documented elevated LAG-3 expression on T cells in settings of with both malignancy and chronic infection (59). PD-1 and LAG-3 are co-expressed on functionally impaired, antigen-specific CD8⁺ T cells (60), and this co-expression has also been observed on both CD4⁺ and CD8⁺ tumor-infiltrating lymphocytes (TILs) in murine cancer models (61). LAG-3 blockade has been shown to augment T cell responses against tumors (62), and concurrent blockade of LAG-3 and PD-1 is more effective than inhibition of either pathway alone (61, 63). Both pathways exert inhibitory effects on T cell responses during chronic viral infection and malignancy. Preclinical

studies demonstrate that dual LAG-3/PD-1 blockade synergistically restores exhausted T-cell function. This mechanistic rationale is supported by clinical evidence from the RELATIVITY-047 trial (NCT03470922), in which nivolumab plus relatlimab improved progression-free survival compared with nivolumab alone in advanced melanoma.

Collectively, the evidence reviewed in this section indicates that LAG-3 has evolved from an emerging immune checkpoint target to a clinically relevant component of cancer immunotherapy, particularly following the demonstrated benefit of dual LAG-3/PD-1 blockade in advanced melanoma. Preclinical studies consistently support the synergistic interaction between LAG-3 and PD-1 blockade, showing enhanced restoration of exhausted T-cell function and improved antitumor immune responses compared with targeting either pathway alone. However, comparison of these findings with clinical observations reveals that the benefit of LAG-3-targeted therapy is not uniform across malignancies. While meaningful activity has been observed in melanoma, studies in several other solid tumors have reported more modest response rates, even in cases with elevated LAG-3 expression. This discrepancy suggests that LAG-3 expression alone may not adequately predict functional dependence on the LAG-3 pathway or therapeutic response. The evidence further indicates that the efficacy of LAG-3 blockade depends on the broader immune context, including the degree of T-cell exhaustion, co-expression of additional inhibitory receptors, and the organization of immune cells within the tumor microenvironment. Therefore, current evidence supports that future development of LAG-3-targeted therapy should emphasize biomarker-guided patient selection and rational combination strategies rather than use of LAG-3 inhibition as monotherapy.

T cell immunoglobulin and ITIM domain (TIGIT)

TIGIT is an inhibitory receptor of the

PVR-like family, expressed on regulatory T cells (Tregs), memory T cells, as well as NK cells (64, 65). It binds two ligands, two ligands, CD112 (also known as Nectin-2, PRR2, or PVRL2) and CD155 (also known as PVR or Nectin-5), which are expressed on antigen presentation cells (APCs), T cells, and non-hematopoietic cells including melanoma cells (66). Notably, CD155 and CD112 are upregulated in several cancers, including melanomas (67). TIGIT outcompetes the costimulatory receptor CD226 (DNAM-1) and CD96 for CD155 binding due to its higher affinity, thereby disrupting CD226-mediated costimulation—a mechanism analogous to CTLA-4 outcompeting CD28 for B7 ligand (2, 9). In TIGIT-deficient mice, tumor progression is significantly slowed (68-70). Additionally, TIGIT indirectly suppresses T cell activation through engagement of CD155 on DCs. This interaction inhibits IL-12 production while promoting IL-10 secretion, fostering tolerogenic DCs that dampen T cell reactions (68).

Studies have focused on TIGIT's role in cancer initiation and progression. Evidence indicates that TIGIT suppresses cytotoxic activity, disrupts granule polarization, and inhibits cytokine production. These effects depend on TIGIT's cytoplasmic immunoreceptor tyrosine-based inhibitory motif (ITIM) and an immunoglobulin tail tyrosine (ITT)-like domain (71). Upon receptor engagement, the ITT-like motif is phosphorylated and recruits the adaptor Grb2, which in turn recruits SHP1 to inhibit phosphoinositide 3-kinase (PI3K) and mitogen-activated protein kinases (MAPKs) signaling (71). Separately, TIGIT associates with β -arrestin 2 to recruit SHP1, impairing TRAF6 autoubiquitination and inhibiting NF- κ B signaling, thereby suppressing interferon- γ (IFN- γ) production (72, 73). TIGIT also dampens TCR signaling, particularly phospholipase C (PLC)-dependent pathways, to suppress IFN- γ and IL-17 production (69, 74). TIGIT upregulates anti-apoptotic molecules such as Bcl-xL and

cytokine receptors for IL-2, IL-7, and IL-15 which support T cell survival (75). Through these mechanisms, TIGIT helps preserve T cell homeostasis while restraining excessive activation (75).

Current evidence reveals fundamental gaps in our understanding of the TIGIT axis. Consensus is lacking regarding the functional significance of nectin-3 as a TIGIT binding partner (64), and the relationship between TIGIT and CD112 requires further investigation. Despite these unanswered questions, TIGIT remains a promising therapeutic target. TIGIT expression is elevated in the tumor microenvironment relative to adjacent peripheral regions, and anti-TIGIT monoclonal antibody may enable targeted immunotherapy with a lower risk of autoimmune adverse effects. Murine studies suggest that TIGIT targeting may be associated with fewer irAEs than anti-PD-1 or anti-CTLA-4 therapy (76). Co-administration of TIGIT inhibitors with anti-PD-1/PD-L1 or anti-TIM-3 agents has demonstrated synergistic suppression of tumor growth (70, 77), although complete remission was not achieved. Preclinical studies demonstrate that TIGIT blockade enhances cytotoxic T-cell and NK-cell activity, particularly in combination with PD-1/PD-L1 inhibition (77). Early clinical evidence from the phase II CITYSCAPE trial (NCT03563716) reported promising antitumor activity for tiragolumab plus atezolizumab in PD-L1-positive non-small cell lung cancer. However, subsequent evaluations have yielded mixed results, indicating that the clinical efficacy of TIGIT-targeted therapy requires further validation.

Collectively, the TIGIT axis exemplifies a broader challenge in immuno-oncology: target expression alone is a poor predictor of functional dependence, and clinical benefit is likely determined by the immunological context rather than by the target itself. Unlike PD-1/PD-L1, TIGIT operates at the interface of T-cell and NK-cell regulation, and its therapeutic impact may depend on the balance between CD226-mediated

costimulation and TIGIT-mediated inhibition, as well as on Fc engineering and the abundance of ligand-expressing cells within the tumor microenvironment. This complexity argues against evaluating TIGIT blockade as a simple substitute for PD-1/PD-L1 inhibition and instead supports context-specific development. Key priorities include functional biomarkers (such as CD226/TIGIT ratios, NK-cell activation states, and spatial co-expression of inhibitory receptors) and prospective incorporation of these variables into trial design. In the broader landscape of resistance to immune checkpoint blockade, TIGIT serves as a paradigm for how a promising preclinical target can be refined through clinically informed, context-dependent strategies rather than dismissed after inconsistent monotherapy results. Ultimately, the clinical value of TIGIT-targeted therapy may depend less on the target itself and more on identifying the right

immunological context in which it matters.

RESISTANCE TO IMMUNE CHECKPOINTS BLOCKADE

Innate or primary resistance

Approximately 60% to 75%, of patients treated with anti-PD-1 checkpoint inhibitors exhibit primary resistance, a proportion that varies by tumor type and disease stage (78). Over 70%, of patients treated with anti-CTLA-4 checkpoint inhibitors similarly show innate resistance (78). Mechanisms underlying primary resistance include reduced infiltration of antitumor T cells (82), lower mutational burden and neo-antigen production (83), and a decreased IFN- γ secretion (84). High PD-L1 expression has been associated with disease progression in some malignancies and can contribute to primary resistance to PD-L1 blockade (85) (Figure 1).

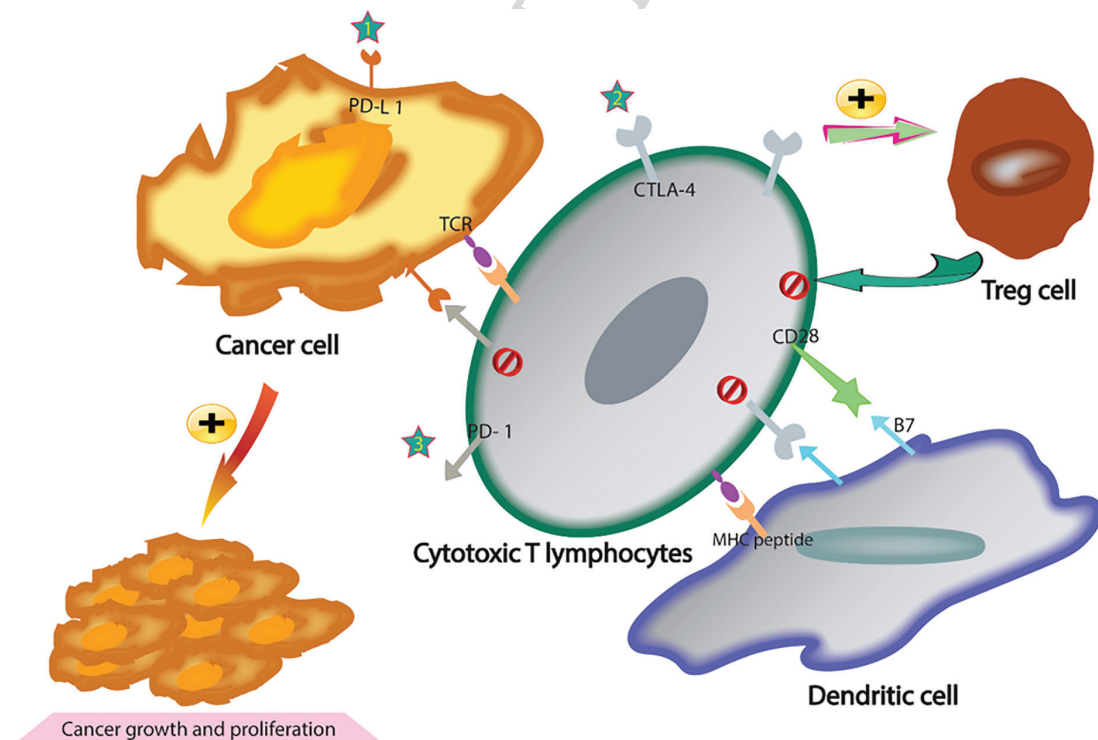


Figure 1. Immune checkpoints blockade in cancer therapy. Inhibition of cytotoxic T cells by regulatory T (Treg) cells and immune checkpoints promotes survival and growth of cancer cells (areas marked with red marks). In addition, CTLA-4 engagement increases proliferation of Treg cells, which further inactivates cytotoxic T cells. Antibody-mediated blockade of immune checkpoint reactivates cytotoxic T cells and induces cancer cells death (areas marked with green stars). PD-L1: programmed death-ligand 1; PD-1: programmed death-1; TCR: T-cell receptor; CTLA-4: cytotoxic T-lymphocyte associated protein 4; MHC: major histocompatibility complex.

Additional host-related factors influence innate resistance, including autoimmune disease, organ or bone marrow transplantation, organ dysfunction, extreme of age (infancy or advanced age), pregnancy, and bacterial, viral, or fungal infections. However, the precise contribution of these host-related determinants remains poorly understood (86, 87).

Collectively, the evidence discussed in this section suggests that primary resistance to immune checkpoint blockade is a multifactorial phenomenon rather than the product of a single molecular alteration. The mechanisms described above—limited T-cell infiltration, low neoantigen availability, impaired interferon signaling, dysregulated checkpoint ligand expression, and host-related immune conditions—likely interact to produce a tumor microenvironment that is insufficiently immunogenic. Their relative contribution vary substantially across tumor types and individual patients, which helps explain the marked variability in clinical response to anti-PD-1 and anti-CTLA-4 therapies. This exposes a key limitation of single-biomarker prediction. Consequently, more comprehensive frameworks integrating tumor molecular features, immune-cell infiltration patterns, cytokine signaling status, and host-related immune characteristics are likely to offer a more reliable basis for identifying patients who may benefit from immune checkpoint blockade.

Acquired or secondary resistance

Patients who initially respond to immune checkpoint blockade but subsequently relapse are considered to have acquired or secondary resistance. The mechanisms underlying acquired resistance may overlap with those of primary resistance though current understanding remains incomplete. Tumor cells can evade the immune detection through genetic alterations, including loss of beta 2 microglobulin (B2M) (88). B2M is an essential subunit of MHC class I molecules, and its loss impairs antigen presentation to CD8⁺ T cells (89). In solid tumors such as

melanoma and lung cancer, acquired B2M loss leads to reduced MHC class I surface expression and diminished CD8⁺ T cell recognition of tumor antigens, contributing to acquired resistance to immune checkpoint blockade (90) (Figure 2).

Mutations in Janus kinase 1 and 2 (JAK1/2) are recognized markers of acquired resistance to immunotherapy. The mutations reduce tumor cell sensitivity to IFN- γ , limiting its antiproliferative effects (91). IFN γ , secreted by activated T cells, plays a central role in modulating immune responses. It activates JAK1/2, leading to phosphorylation of tyrosine residues on signal transducer and activator of transcription (STAT) proteins. The JAK/STAT pathway governs diverse biological processes, including cell motility, differentiation, and proliferation. IFN γ also induces PD-L1 upregulation on cancer cells, suppressing anti-tumor T lymphocytes (92). In patients with acquired resistance, tumor cells harbor loss-of-function mutations in the JAK1 and JAK2, abrogating IFN- γ signaling. These patients initially responded to pembrolizumab therapy, but responses diminished after a median of approximately 1.8 years (91).

Tumor cells can also develop resistance in response to changes in their surrounding microenvironment (88). For example, cytokine stimulation—particularly IFN γ released by T lymphocytes—upregulates PD-L1 on tumor cells. High PD-L1 expression then impairs antitumor T cell activity, reducing tumor suppression (93). Both primary and acquired resistance can involve this mechanism (88). Acquired resistance can also arise from the compensatory upregulation of additional immune checkpoints, particularly LAG-3 and TIM-3, which further suppress T-cell function (92). A recent study found that individuals who developed resistance to anti-PD-1 immunotherapy showed increased TIM-3 expression (94).

Collectively, the evidence discussed in this section suggests that acquired resistance to immune checkpoint blockade is a dynamic

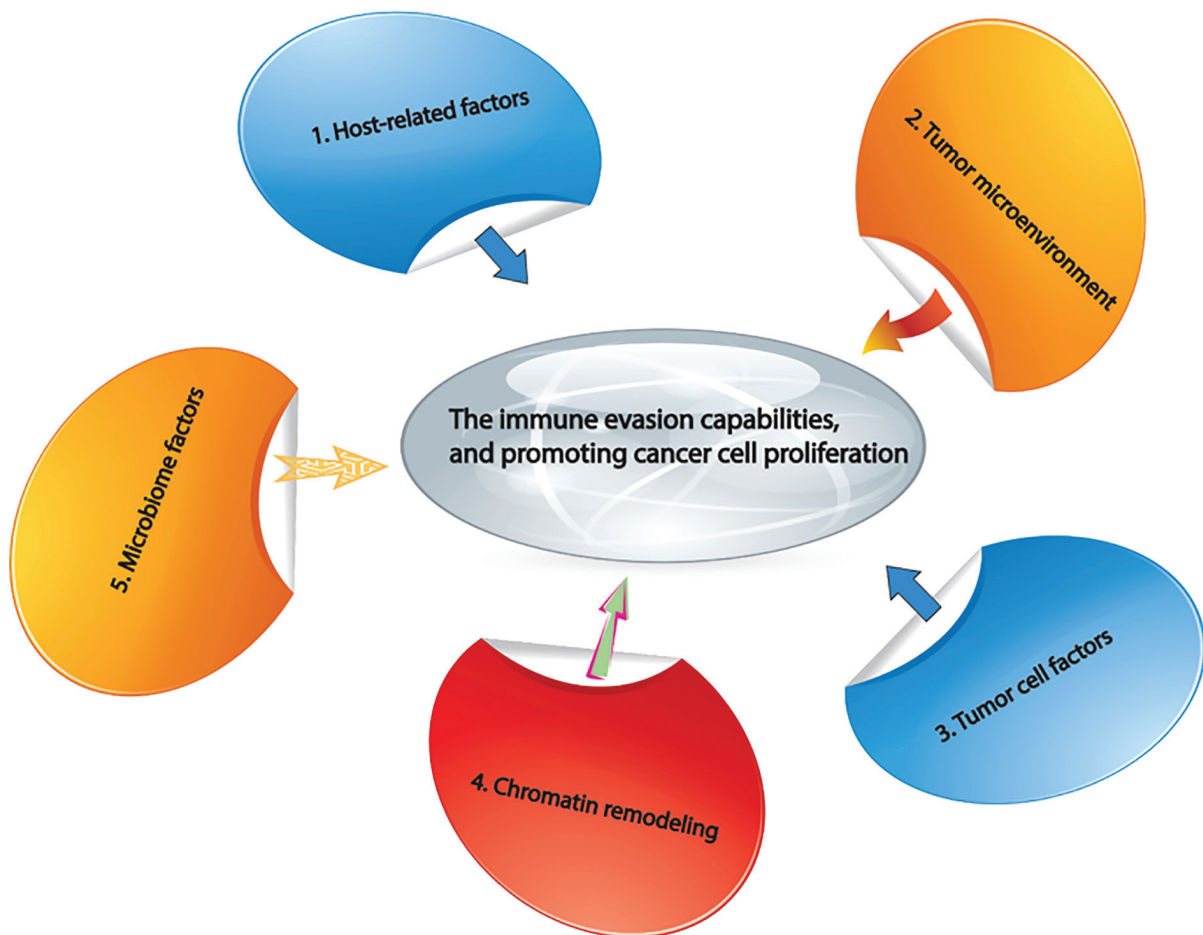


Figure 2. The mechanisms of resistance to monoclonal antibody-mediated immune checkpoint blockade.

process that evolves under therapeutic selective pressure. Unlike primary resistance, which often reflects pre-existing tumor and immune microenvironment characteristics, acquired resistance emerges after an initial clinical response and reflects adaptive changes that enable immune evasion. The mechanisms described above—altered antigen presentation, impaired interferon signaling, increased inhibitory ligand expression, and compensatory checkpoint activation—are not mutually exclusive and may occur simultaneously or sequentially during disease progression. Importantly, treatment-induced immune activity can itself reshape the tumor microenvironment, promoting conditions that favor immune escape. This evolving interplay between tumor-intrinsic alterations and adaptive immune responses helps explain why patients who initially benefit from checkpoint inhibition may later

relapse. Consequently, effective management of acquired resistance will likely require longitudinal monitoring of tumor and immune changes, alongside therapeutic strategies capable of targeting multiple complementary resistance mechanisms.

RESISTANCE FACTORS IN IMMUNE CHECKPOINTS BLOCKADE

Resistance to immune checkpoint blockade is a multifactorial process that reflects interactions among tumor biology, the tumor microenvironment, and host-related determinants. To organize this evidence, resistance mechanisms can be categorized into three interrelated groups: tumor-intrinsic factors, tumor-extrinsic (microenvironmental) factors, and host-related (systemic and germline) factors. This framework facilitates

a systematic interpretation of the current evidence and highlights the interconnected biology underlying response to ICB. Although these categories are presented separately, they are highly interconnected and collectively shape therapeutic outcomes (95).

Host-related resistance mechanisms

Host-related resistance mechanisms involve systemic factors that influence immune competence and modulate responses to ICB. Numerous studies have investigated how host-related characteristics affect outcomes following immune checkpoint inhibition (95). These include circulating levels of lactate dehydrogenase (LDH), C-reactive protein (CRP), eosinophils, monocytes, neutrophils, platelets, and lymphocytes (96, 97). Lower LDH levels have been linked to more favorable treatment responses (98). Increases in plasma soluble PD-1 (sPD-1) during anti-PD-1 therapy have been correlated with tumor progression, suggesting sPD-1 as an early biomarker of primary resistance (99).

Peripheral blood cell counts have also been linked to clinical outcomes. Elevated neutrophil and platelet counts are generally associated with accelerated cancer progression, whereas higher lymphocyte counts correlate with delayed disease progression (100). Following treatment with nivolumab or pembrolizumab, elevated neutrophil and platelet counts have been associated with a higher risk of disease progression, as reviewed by Seto et al (101). These findings suggest that host-related circulating biomarkers may provide clinically relevant information for predicting immunotherapy outcomes.

The gut microbiome is another host-related factor influencing responsiveness to ICB. Greater microbial diversity and enrichment of specific bacterial taxa are associated with enhanced immune responses to checkpoint blockade (102, 103). Germ-free mice receiving fecal transplants from patients who responded favorably to ICB demonstrated improved outcomes when treated with anti-PD-L1 therapy (104). Conversely, antibiotic

exposure can alter microbiome composition and potentially reduce ICB effectiveness (105, 106). The association between gut microbiota composition and immune response to ICB is partly explained by cross-reactivity between microbial and tumor antigens, which enhance dendritic-cell activation, increase antigen presentation, and stimulate inflammatory cytokine production (107, 108).

Collectively, the evidence suggests that host-related resistance mechanisms contribute substantially to the heterogeneity of responses observed with immune checkpoint inhibitors. Circulating biomarkers, peripheral immune-cell profiles, and gut microbiome composition each provide insight into the host's systemic immune status and its capacity to support antitumor immunity. However, the predictive value of these factors is not uniform. Elevated neutrophil and platelet counts generally associate with poorer clinical outcomes, while higher lymphocyte counts associate with more favorable responses, but the strength and consistency of these associations vary across patient populations, tumor types, and treatment settings. Similarly, although increased microbial diversity and specific bacterial taxa have been linked to improved response, the microbiome signatures identified in responders are not consistently replicated across independent studies. These discrepancies likely reflect the dynamic and context-dependent nature of host immunity, which is shaped by systemic inflammation, metabolic status, prior therapies, antibiotic exposure, and environmental conditions. Host-related factors should therefore be interpreted as interacting and evolving contributors to immunotherapy responsiveness rather than as isolated predictive biomarkers. Future patient stratification will likely require integrated models combining circulating biomarkers, immune-cell profiling, microbiome characteristics, and relevant clinical variables.

Tumor-extrinsic resistance mechanisms

Tumor-extrinsic resistance mechanisms are mediated by components of the tumor

microenvironment that suppress antitumor immunity and limit the effectiveness of ICB. The tumor microenvironment comprises a complex network of cancer cells, extracellular matrix, stromal cells, immune cells, cytokines, and metabolic mediators that collectively shape immune responsiveness (109). It sustains chronic inflammation, suppresses immune responses, promotes angiogenesis, facilitates tumor cell proliferation, and enhances immune evasion (110). PD-L1 engagement within the tumor microenvironment further contributes to immune evasion (111). Tumor microenvironments that respond to or resist anti-PD-1 therapy may have similar CD8⁺ T-cell densities, but differences in other immune-cell populations and suppressive mechanisms can substantially influence treatment outcomes.

Altered regulatory T cell (Treg) frequency within the tumor microenvironment is associated with resistance to ICB across several cancer types (79). Altered regulatory T cell (Treg) frequency within the tumor microenvironment (TME) is associated with resistance to immune checkpoint blockade across several cancer types (79). Other immunosuppressive populations, including myeloid-derived suppressor cells (MDSCs), tumor-associated macrophages (TAMs), and cancer-associated fibroblasts (CAFs), also inhibit cytotoxic T-cell function and promote tumor progression.

Metabolic mechanisms within the tumor microenvironment further contribute to resistance. Indoleamine 2,3-dioxygenase (IDO), expressed in myeloid and tumor cells, degrades tryptophan into immunosuppressive metabolites such as kynurenine (112). Tryptophan depletion and metabolite accumulation can induce T cells apoptosis and impair antitumor immunity. Preclinical studies have shown that IDO inhibitors combined with immune checkpoint inhibitors enhance T-cell activity and promote eradication of IDO-expressing tumors (113). However, clinical translation has been challenging. In the phase III ECHO-

301/KEYNOTE-252 trial (NCT02752074), epacadostat plus pembrolizumab did not improve progression-free survival compared with pembrolizumab alone in advanced melanoma (114). This outcome likely reflects multiple factors—including uncertain target engagement at the dose tested, patient selection bias, and lack of biomarker-based enrollment—and underscores the need for rigorous clinical validation of preclinical resistance mechanisms.

Collectively, the evidence indicates that tumor-extrinsic resistance arises from a coordinated network of suppressive factors within the tumor microenvironment rather than from a single dominant mechanism. Although CD8⁺ T cells infiltration is often considered a favorable indicator of response to ICB, outcomes also depend strongly on the functional state of these cells and on the abundance of immunosuppressive populations such as Tregs, MDSCs, TAMs, and CAFs, as well as metabolic alterations including IDO-mediated tryptophan degradation and kynurenine accumulation. This explains a key contradiction in the literature: tumors with apparently similar immune-cell infiltration patterns may respond very differently because of differences in suppressive cellular interactions, cytokine signaling, and metabolic conditions within the tumor microenvironment. Furthermore, while preclinical studies targeting individual tumor-microenvironment components have shown encouraging results, their clinical translation has been inconsistent—a discrepancy that likely reflects the interconnected and compensatory nature of tumor-extrinsic resistance pathways. Current evidence therefore supports integrated therapeutic approaches that simultaneously target immune checkpoint pathways, suppressive cellular populations, and metabolic mechanisms to overcome resistance and improve long-term clinical outcomes.

Tumor-intrinsic resistance mechanisms

Tumor-intrinsic resistance mechanisms

arise from genetic, epigenetic, and signaling alterations within cancer cells that impair immune recognition and reduce sensitivity to ICB. Several proteins and signaling pathways have been implicated in this form of resistance.

Activation of the WNT/ β -catenin signaling pathway correlates with the absence of T cells and CD103⁺ dendritic cells within the tumor microenvironment. This pathway suppresses CCL4 production and confers resistance to monoclonal antibodies targeting PD-L1 and CTLA-4 (115). Loss of phosphatase and tensin homolog (PTEN) and activation of the PI3K/AKT cascade similarly contribute to resistance (116).

In Hodgkin's lymphoma, where genetically driven PD-L1 overexpression confers sensitivity to PD-1 blockade, epidermal growth factor receptor (EGFR) activation upregulates PD-L1 alongside PD-1, CTLA-4, and inflammatory cytokines, creating a broader immunosuppressive program that contributes to resistance (117).

The efficacy of ICB also depends on T cell activation against tumor neoantigens. Consequently, tumor mutational burden is a key determinant of responsiveness to ICB (118). Elevated mutational burden, nonsynonymous mutations, and microsatellite instability are associated with improved responses to PD-1/PD-L1 blockade across several malignancies (119).

Chromatin remodeling represents an additional tumor-intrinsic mechanism influencing sensitivity to ICB. SWI/SNF chromatin-remodeling complexes are frequently mutated in cancer, with subunit mutations identified in approximately 20% of all human tumors (120, 121). The PBAF complex (ARID2, PBRM1, and BRD7), and the BAF complex (ARID1A/B subunits) are particularly relevant (121). PBRM1 loss disrupts normal PBAF function and increases tumor sensitivity to PD-1 blockade (122). Mechanistically, PBAF subunit loss—particularly of PBRM1—increases chromatin accessibility at IFN-

inducible gene loci, promoting CXCL9/CXCL10 production and T cell recruitment (122). In clear cell renal cell carcinoma, PBRM1 loss has been associated with increased clinical benefit from ICB, though this finding remains context-dependent (123). ARID1A deficiency impairs mismatch repair, through disrupted MSH2 recruitment, increasing tumor mutational burden and microsatellite instability, and heightens sensitizes tumors to PD-L1 inhibition (124).

Collectively, the evidence indicates that tumor-intrinsic resistance to ICB is driven by a complex interplay of genetic, epigenetic, and signaling alterations that impair immune recognition and antitumor immune activation. Pathways such as WNT/ β -catenin activation, PTEN loss, PI3K/AKT signaling, JAK/STAT-mediated PD-L1 upregulation, EGFR signaling, and chromatin-remodeling abnormalities can each contribute to immune evasion, but their relative importance varies substantially across tumor types and biological contexts. Notably, some alterations—including elevated tumor mutational burden, microsatellite instability, and specific chromatin-remodeling changes—are associated with improved responsiveness to PD-1/PD-L1 blockade, whereas comparable alterations do not consistently predict therapeutic benefit in other settings. This variability exposes a key limitation of single-molecular alterations as biomarkers. Furthermore, many tumor-intrinsic pathways with strong mechanistic associations in experimental studies have not uniformly translated into durable clinical benefit, in part because tumor-intrinsic alterations interact with microenvironmental and host-related resistance mechanisms. Therefore, current evidence supports integrated molecular and immune-context analyses—combining genomic profiling, epigenetic characterization, and tumor microenvironment assessment—to improve patient stratification and guide rational combination therapies.

RESISTANCE MECHANISMS IN IMMUNE CHECKPOINTS BLOCKADE

The precise mechanisms underlying resistance to ICB remain incompletely understood. Our knowledge of the molecular and clinical variables that shape response to PD-1, PDL-1, and CTLA-4 blockade, is still limited (125, 126). It is well established, however, that activating and expanding antitumor T cells within the tumor microenvironment is essential (127). CD8⁺ T cell activation requires tumor-associated antigen presentation by APCs and DCs, followed by recognition of MHC class I molecule. T cell receptors (TCRs) recognize tumor antigens presented on MHC molecules, initiating the primary signal required for T cell activation (111). CD8⁺ T cells then mature

into effector T cells and undergo clonal proliferation before migrating into the tumor microenvironment (127). To ensure durable immune responses, CD4⁺ T cells and DCs promote the transition of a subset of effector T cells into effector memory T cells (128), establishing an immune response capable of recognizing and responding to the antigen upon re-exposure (129). The limited efficacy of ICB can be attributed to defects in this process, which fall into three categories: inadequate generation of antitumor T cells, impaired functionality of antitumor T cells, and insufficient development of memory T cells (127).

Multiple mechanisms have been proposed to predict resistance to ICB (Figure 3). Antigen presentation plays a critical role in this process. Deletion or alteration of B2M

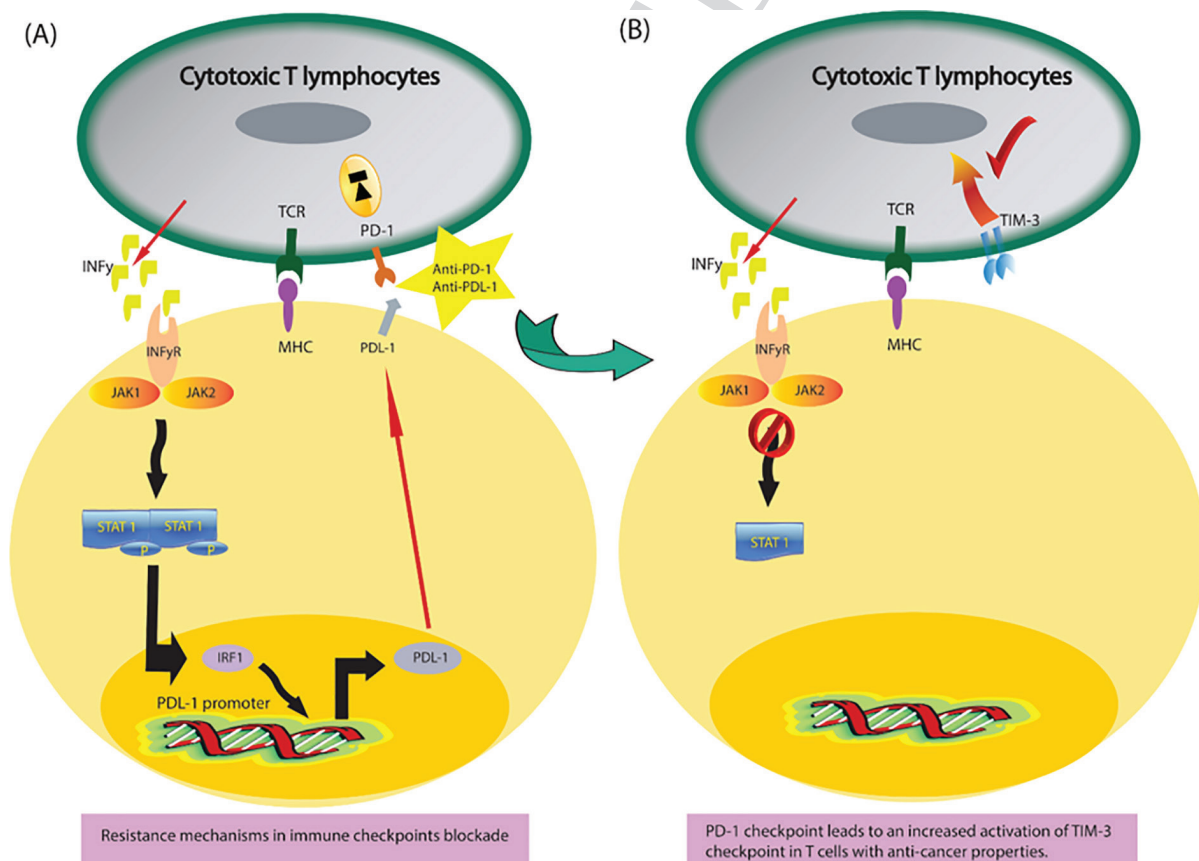


Figure 3. Mechanisms of resistance to immune checkpoint blockade and their therapeutic reversal. (A) Interferon-gamma (IFN γ) secreted by activated cytotoxic T cells induces PD-L1 on cancer cells. PD-L1 binding to PD-1 on cytotoxic T cells transmits an inhibitory signal, leading to T cell inactivation and immune evasion. (B) Anti-PD-L1 and anti-PD-1 block this interaction, restoring cytotoxic T cell activity and promoting antitumor immunity. TCR: T-cell receptor; PD-L1: programmed death-ligand 1; PD-1: programmed death-1; MHC: major histocompatibility complex; IFN- γ : interferon gamma; TIM-3: T-cell immunoglobulin and mucin domain 3.

and human leukocyte antigen (HLA) genes class I genes — which encode components of the MHC class I antigen-presentation machinery — impairs immune cell activation (130). Inadequate antigen presentation by tumor cells consequently hinders cytotoxic T lymphocyte responses. In addition, B2M mutations have been associated with primary resistance to ICB (131).

JAK1 and JAK2 mutations are established drivers of both primary and acquired resistance to ICB (91, 132). Homozygous PTEN deletion represents another contributing mechanism. Loss of PTEN is associated with resistance to anti-PD-1 therapy (133). Downstream, reduced CCL4—a cytokine required for DC activation — decreases the number of antitumor T cells in the tumor microenvironment. This reduction reflects activation of Wnt/ β -catenin signaling in cancer cells, which suppresses CCL4 production and T cell recruitment (115).

Immunosuppressive cells—particularly MDSCs and Treg cells—together with elevated levels of immunosuppressive mediators such as vascular endothelial growth factor (VEGF), impair T cell function and limit tumor eradication (134, 135). MDSCs are a heterogeneous myeloid progenitor (136) that impair cytotoxic T cell activity. Circulating MDSCs may serve as a prognostic factor for resistance to immune checkpoint inhibitors (137), and prior studies have shown that ICB can itself drive myeloid cells expansion (138, 139). Tregs are a distinct subset of CD4⁺ T cells with immunosuppressive properties, defined by expression of the transcription factor FoxP3 (140, 141). They suppress CD8⁺ T cell cytotoxicity through multiple mechanisms, including reduced IL2 production, secretion of immunosuppressive cytokines such as IL-10 and transforming growth factor beta (TGF- β), and upregulation of CTLA-4 (142). Treg, MDSCs, and other immunosuppressive populations such as M2 TAMs are considered viable targets for overcoming ICB resistance (143).

Tumor cells frequently upregulate

alternative immune checkpoints, contributing to resistance to ICB (144). In murine lung cancer model, PD-1 blockade increased TIM-3 expression on antitumor T cells, correlating with tumor progression after an initial response (94). Aneuploidy represents another mechanism of resistance. Aneuploid tumors show reduced cytotoxic T lymphocyte infiltration, a known contributor to immunotherapy failure, and aneuploidy has been recognized as a significant driver of ICB resistance (145).

Epithelial-to-mesenchymal transition (EMT), through which epithelial tumor cells acquire mesenchymal and stem-like properties, is widely recognized as central to cancer initiation and progression (146). Within this pathway, 147 genes have been linked to innate resistance to PD-1 blockade. Upregulation of mouse double minute 2 (MDM2) and MDM4 proteins inhibits TP53 activity, driving resistance to PD-1 blockade. However the specific role of this pathway in patients receiving anti-PD-1 therapy remains unclear (147, 148).

The gut microbiome also substantially influences ICB response, though the mechanisms underlying this interaction are incompletely understood. Reduced microbial diversity is associated with increased ICB resistance, and antibiotic administration prior to therapy exacerbates this effect (149, 150).

Collectively, the evidence reviewed in this section indicates that resistance to ICB is a multifactorial and dynamic process that cannot be attributed to a single molecular alteration or immune pathway. The contribution of each mechanism described above varies by tumor type, disease stage, treatment context, and the evolving interaction between tumor cells and the immune microenvironment. While several mechanisms have shown strong experimental associations with resistance, their predictive value and therapeutic relevance have not been consistently confirmed in clinical settings. Notably, primary and acquired resistance appear to arise through distinct but overlapping processes: primary resistance is

often associated with pre-existing immune exclusion, whereas acquired resistance develops through immunoediting, clonal selection, and compensatory activation of alternative inhibitory pathways. These observations help explain why strategies targeting single resistance pathways have generally produced limited or variable clinical benefit. Therefore, current evidence supports integrated approaches that combine molecular profiling, immune monitoring, and rational combination therapies to address the heterogeneous and evolving mechanisms of ICB resistance.

TRANSLATIONAL RELEVANCE AND FUTURE CLINICAL OPPORTUNITIES

Understanding resistance mechanisms to ICB has important implications for the developing precision immunotherapy strategies and designing future clinical trials. The classification of resistance mechanisms into tumor-intrinsic, tumor-extrinsic, and host-related factors provides a practical framework for identifying biomarkers that may improve patient stratification and guide treatment selection. By integrating these mechanistic insights may enhance the predictive accuracy of immunotherapy response and reduce exposure to ineffective treatments (151, 152). Although immune checkpoint inhibitors have transformed the treatment landscape of several malignancies, clinical benefit remains highly heterogeneous across tumor types and individual patients. Recent phase III trials, including CheckMate 067 (NCT01844505), CheckMate 141 (NCT02105636), KEYNOTE-040 (NCT02252042), and RELATIVITY-047 (NCT03470922), have demonstrated meaningful survival benefits in selected settings. Nevertheless, a substantial proportion of patients exhibit primary resistance or develop acquired resistance after an initial response, underscoring the need for biomarker-guided patient selection and rational combination strategies.

Tumor-intrinsic biomarkers represent a major opportunity for patient stratification. Antigen presentation defects, such as $\beta 2M$ loss and HLA alterations, impair T-cell recognition of tumor cells and may identify patients with reduced likelihood of responding to immune checkpoint inhibitors (153). Similarly, high tumor mutational burden and microsatellite instability have been associated with increased neoantigen generation and improved responsiveness to PD-1/PD-L1 blockade in selected malignancies (154, 155). Other molecular alterations—including WNT/ β -catenin activation, PTEN loss, PI3K/AKT signaling, and JAK/STAT-mediated PD-L1 upregulation—may further refine biomarker-based selection by identifying tumors with distinct immune-evasion profiles (156, 157).

Tumor-extrinsic biomarkers within the tumor microenvironment also hold significant translational value. PD-L1 expression remains one of the most widely used biomarkers for selecting patients for PD-1/PD-L1-targeted therapies; however, its predictive performance varies across tumor types and clinical settings (158). The density and spatial distribution of tumor-infiltrating lymphocytes, particularly CD8⁺ T cells, are associated with improved response to ICB and may complement PD-L1 assessment in future stratification strategies (159). In addition, the abundance of regulatory T cells, MDSCs, TAMs, and immunosuppressive cytokines such as TGF- β and IL-10 may inform resistance likelihood and potential benefit of combination therapies targeting the tumor microenvironment (160, 161).

Host-related factors are increasingly recognized as valuable sources of predictive biomarkers. Circulating biomarkers—including lactate dehydrogenase, C-reactive protein, neutrophil-to-lymphocyte ratio, and peripheral immune-cell profiles—are associated with outcomes in patients receiving ICB (162, 163). The gut microbiome has emerged as a particularly promising host-related biomarker, as specific compositions and higher diversity have been linked to

enhanced antitumor immune responses and improved responsiveness to PD-1/PD-L1 blockade across several studies (164, 165). Although microbiome-based interventions remain investigational, these findings support the incorporating microbiome analysis into future biomarker-driven clinical trials.

The mechanistic insights summarized here provide a rationale for combination strategies to overcome ICB resistance. Such approaches may pair immune checkpoint inhibitors with targeted agents against oncogenic signaling, anti-angiogenic agents, epigenetic modulators, metabolic inhibitors such as IDO inhibitors, and therapies designed to remodel the tumor microenvironment (166-168). Microbiome-directed interventions, including fecal microbiota transplantation and probiotic or dietary modulation, are also being explored to enhance systemic antitumor immunity and improve responsiveness to immunotherapy (169).

Future clinical trials should therefore adopt biomarker-driven designs incorporating molecular profiling, spatial characterization of the tumor microenvironment, host immune profiling, and longitudinal monitoring of response. Such approaches may reveal dynamic resistance mechanisms, enable adaptive strategies, and improve the likelihood of durable clinical benefit. Integrating tumor-intrinsic, tumor-extrinsic, and host-related biomarkers into trial design could ultimately advance precision immunoncology and translate mechanistic insights into more effective, individualized therapies for patients with cancer (170, 171).

CURRENT CHALLENGES AND FUTURE DIRECTIONS

Although immune checkpoint inhibitors (ICIs) have transformed the therapeutic landscape of oncology by inducing durable antitumor responses in selected malignancies, their clinical benefit remains highly heterogeneous across tumor types and individual patients

(172, 173). Clinical trials and translational studies have demonstrated substantial improvements in overall survival and disease control in selected settings, particularly with agents targeting the CTLA-4 and PD-1/PD-L1 pathways (174). However, comparison across studies reveals marked variability in objective response rates, progression-free survival, and durability of benefit, with a considerable proportion of patients exhibiting primary resistance or developing acquired resistance following an initial therapeutic response (79). This underscores a fundamental limitation of current immunotherapy paradigms: therapeutic efficacy is determined not by checkpoint expression alone but by the dynamic interaction among tumor-intrinsic genetic alterations, epigenetic regulation, antigen presentation capacity, the immunosuppressive tumor microenvironment, and host immune competence (79). Furthermore, while combination regimens involving ICIs and other therapeutic modalities have demonstrated enhanced antitumor activity in some preclinical and clinical contexts (172, 173), their clinical translation has been constrained by increased immune-related toxicity, variable efficacy across patient populations, and the absence of universally reliable predictive biomarkers. Consequently, future progress in ICB will require the integration of comprehensive molecular profiling, immune-contexture analysis, and rational biomarker-guided combination strategies to identify patients most likely to benefit from therapy and to overcome the multifactorial mechanisms underlying immune resistance.

CONCLUSION

Immune checkpoint blockade (ICB) has revolutionized cancer treatment by providing durable clinical benefits in a subset of patients with diverse malignancies. However, accumulating evidence demonstrates that these successes are accompanied by considerable

biological and clinical heterogeneity, as many patients exhibit either primary resistance or develop acquired resistance during treatment. Critical evaluation of evidence indicates that resistance cannot be attributed to a single molecular mechanism or immune checkpoint. Instead, it arises from dynamic interactions among tumor-intrinsic genetic and epigenetic alterations, the tumor microenvironment, host-related factors, immune-cell plasticity, and adaptive evolutionary processes. Although numerous preclinical studies have identified promising targets, their translation into practice has frequently yielded inconsistent or modest outcomes, highlighting the limitations of current experimental models and single-target strategies. Likewise, predictive biomarkers such as PD-L1 expression, tumor mutational burden, and microsatellite instability have shown variable performance across different tumor types, emphasizing the need for more comprehensive and context-dependent approaches to patient stratification. Collectively, current evidence supports a shift from reductionist models of immune resistance toward integrated precision immuno-oncology strategies. Future advances will likely depend on combining multi-omics technologies, spatial and single-cell profiling, longitudinal immune monitoring, and robust biomarker-guided patient selection to better characterize resistance mechanisms and optimize decision-making. Rational combinations targeting multiple complementary pathways are expected to improve clinical translation, overcome resistance, and expand the proportion of patients who achieve durable responses to immune checkpoint blockade.

ABBREVIATIONS

PD-L1: programmed death-ligand 1; PD-1: programmed cell death protein-1; CTLA-4: cytotoxic T-lymphocyte antigen 4; LAG-3: lymphocyte activation gene 3; KIR: killer immunoglobulin-like receptors; TIM-3:

T cell immunoglobulin mucin 3; TILs: tumor-infiltrating leukocytes; NK: natural killer; NSCLC: non-small-cell lung cancer; scRNA-seq: single-cell RNA sequencing; HNSCC: head-and-neck squamous cell carcinoma; OS: overall survival; irAEs: immune-related adverse events; IFN: interferon; DCs: dendritic cells; HIV-1: human immunodeficiency virus 1; HBV: hepatitis B virus; HCV: hepatitis C virus; PBMCs: peripheral blood mononuclear cells; MHC: histocompatibility complex; ITIM: immunoreceptor tyrosine-based inhibitory motif; PI3K: phosphoinositide 3-kinase; MAPKs: mitogen-activated protein kinases; JAK1: Janus kinase 1; STAT: signal transducer and activator of transcription; LDH: lactate dehydrogenase; CRP: C-reactive protein; IDO: indoleamine 2,3-dioxygenase; CCL4: carbon tetrachloride; PTEN: phosphatase and tensin homologue; EGFR: epidermal growth factor receptor; ARID2: AT-rich interaction domain 2; PBRM1: polybromo-1; BRD7: bromodomain-containing protein 7; HLA: human leukocyte antigen; MDSCs: myeloid-derived suppressor cells; VEGF: vascular endothelial growth factor; TGF- β : transforming growth factor beta; MDM2: mouse double minute 2.

AUTHORS' CONTRIBUTION

Y.X. drafted the manuscript and prepared the figures. H.S. conceptualized and supervised the work and critically reviewed and edited the manuscript. Both authors read and approved the final manuscript.

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The authors declare no conflicts of interest.

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