J Bionic Mem 2026; 6(1): 31-51 DOI: 10.53545/jbm.2026.55

Journal of Bionic Memory Review article

Engineering the tumor microenvironment: Mechanisms and technology-enabled strategies to restore anti-tumor immunity

Mustafa Ersoy *

Department of Internal Medicine Sciences, Division of Medical Oncology, Faculty of Medicine, Kütahya Health Sciences University, Kütahya, Türkiye

ABSTRACT

The clinical efficacy of cancer immunotherapy is determined not merely by tumor-intrinsic genetics but by the ecosystem in which malignant cells reside. This ecosystem, the tumor microenvironment (TME), behaves as a dynamic organ composed of stromal networks, aberrant vasculature, extracellular matrix, and immune infiltrates that co-evolve under therapeutic pressure. Although immune checkpoint inhibitors have transformed the management of multiple solid tumors, primary and acquired resistance remain common and typically reflect layered barriers rather than a single defect. These barriers include limited immune recognition from disrupted antigen presentation or inflammatory signaling, physical and chemical exclusion driven by fibroblast programs and vascular dysfunction, suppressive myeloid and regulatory circuits that constrain effector function, and metabolic hostility created by hypoxia, acidosis, and nutrient competition. In this review, we synthesize major immune escape mechanisms across tumor types and organize them into actionable domains that can be matched to therapeutic "modules," including priming approaches, stromal and vascular remodeling, myeloid reprogramming, metabolic checkpoint targeting, and next-generation inhibitory receptor strategies. We also discuss technology-enabled profiling—multiplex imaging, spatial and single-cell profiling, circulating biomarkers, and quantitative digital pathology—that can map immune architecture and functional states with spatial and temporal resolution. Linking mechanism-based models of immune escape with scalable measurement tools may support biomarker-guided combinations and adaptive immunotherapy strategies capable of reprogramming the tumor immune microenvironment to restore durable anti-tumor immunity.

Keywords: Tumor microenvironment, immune escape, immunotherapy resistance, spatial profiling, digital pathology.

*Mustafa Ersoy **

Department of Internal Medicine Sciences, Division of Medical Oncology, Faculty of Medicine, Kütahya Health Sciences University, Kütahya, Türkiye

E-mail: mustafa.ersoy@ksbu.edu.tr

Received: 2026-02-15

Accepted: 2026-03-01 / Published: 2026-03-02

Introduction

Modern oncology has moved from a tumor-cell-only perspective to a systems view in which the tumor microenvironment (TME) is a major determinant of therapeutic outcome [1]. The immune-centered component of the TME—often termed the tumor immune microenvironment (TIME)—is not only a count of infiltrating leukocytes. It also reflects where immune cells reside relative to malignant nests, whether they are activated or dysfunctional, and which stromal, vascular, and metabolic cues shape their behavior [2]. In some tumors, the TIME supports effective priming, trafficking, and cytotoxic activity. In others, the same

immune machinery is diverted into tolerance, paralysis, or spatial exclusion [3].

Immune checkpoint inhibitors (ICIs) have produced durable responses in melanoma, non-small cell lung cancer, renal cell carcinoma, and several additional malignancies [4]. Yet, for most tumor types and most patients, checkpoint blockade alone is insufficient. Primary non-response is common, and acquired resistance can develop after an initial response. Translational work increasingly suggests that resistance is rarely explained by a single pathway. Instead, tumors assemble a layered defense in which failures of immune recognition coexist with barriers to immune access and with local suppression [5]. Physical exclusion by fibroblast-rich stroma and dysfunctional vasculature can prevent T-cell contact; suppressive myeloid cells and regulatory T cells dampen effector programs; and hypoxia, acidosis, and nutrient competition erode immune fitness even when infiltration is present [6].

Clinical shorthand often classifies tumors as inflamed, excluded, or desert. This classification is useful because it maps to recurring patterns of failure, but it also compresses mechanistic heterogeneity [7]. An inflamed tumor may be dominated by compensatory inhibitory receptors, by myeloid-derived suppression, or by adenosine-rich hypoxic niches [8]. Exclusion can be driven primarily by fibroblast activation and matrix stiffness in one patient and by dysfunctional vessels and high interstitial pressure in another [9]. A practical review therefore needs to connect phenotype to mechanism, because the choice of "next therapy" depends on which barrier is dominant.

We are now entering an era in which the TME can be approached as something that can be deliberately remodeled. "Engineering" in this context refers to mechanism-based interventions that change access, cell states, and tissue constraints so that anti-tumor immunity can operate [10]. In parallel, measurement platforms with spatial and state resolution are transforming the TIME from a descriptive concept into a set of testable hypotheses [11]. This review organizes immune escape into actionable domains, describes therapeutic strategies that target each domain, and discusses profiling technologies that can map immune architecture and functional states. The overarching aim is to connect what is measurable to what is actionable, and to outline a logic for assembling combinations and adapting therapy over time.

Overcoming Cellular Barriers: Exclusion and Suppression

Immune escape is often enforced by the cellular architecture of TIME. Across many solid tumors, the balance between effector populations (tumor-reactive CD8+ T cells, helper T-cell subsets, and natural killer cells) and suppressive populations (tumor-associated macrophages, myeloid-derived suppressor cells, and regulatory T cells) correlates with prognosis and likelihood of response to immunotherapy [12]. Importantly, these populations are not independent variables: stromal and metabolic cues shape their recruitment and state, and therapeutic pressure can shift the balance rapidly. A mechanistic view therefore treats the cellular landscape as a dynamic circuit rather than as a static list of cell types [13].

The myeloid shield: from suppression to reprogramming

Myeloid cells frequently form the dominant suppressive backbone of the TIME. Tumor-associated macrophages (TAMs) and myeloid-derived suppressor cells (MDSCs) can suppress T-cell priming and effector function through

multiple, partially redundant mechanisms [14]. These include secretion of inhibitory cytokines such as IL-10 and TGF-beta, depletion of key nutrients (for example arginine via arginase-1 and tryptophan via indoleamine 2,3-dioxygenase), production of reactive oxygen and nitrogen species that damage signaling pathways, and expression of inhibitory ligands that reinforce dysfunctional states [15]. Myeloid cells also shape tissue structure by promoting angiogenesis, matrix remodeling, and local chemokine landscapes that favor suppressive recruitment [16].

A key clinical implication is that a tumor can appear immune-rich by bulk measures while remaining resistant because the inflammatory context is dominated by suppressive myeloid programs [17]. Myeloid lineages are also highly plastic. Hypoxia, lactate, colony-stimulating factors, and therapy-induced stress can shift myeloid states over time, contributing to both primary and acquired resistance. This plasticity creates therapeutic opportunity, but it also means that simple depletion has often produced limited benefit due to compensatory recruitment and repopulation. As a result, current strategies increasingly focus on changing function rather than just reducing numbers [18].

Therapeutic strategies have therefore shifted toward functional reprogramming and selective pathway modulation. Approaches targeting colony stimulating factor-1 receptor (CSF-1R) aim to reduce macrophage survival signals and remodel myeloid composition. Agonists of CD40 and related pathways can enhance antigen presentation and promote an immunostimulatory innate compartment that supports T-cell priming and expansion. Additional strategies aim to block myeloid recruitment (for example by interrupting chemokine pathways), inhibit suppressive enzymes, or engage innate phagocytosis programs through macrophage checkpoint modulation. In practice, myeloid-directed interventions are often best positioned as combination partners: they can lower suppressive tone, improve antigen presentation, and increase the probability that inhibitory receptor blockade translates into effective killing [19].

Regulatory T cells and suppressive lymphoid circuits

Regulatory T cells (Tregs) are essential for limiting autoimmunity, but within tumors they often suppress effector immunity and support immune escape. Tregs can inhibit dendritic cell maturation, dampen co-stimulation, consume local IL-2, and contribute to an inhibitory cytokine milieu. They also intersect with myeloid suppression and with metabolic checkpoints such as adenosine, creating reinforcing loops that can be difficult to break with a single agent [20].

The clinical importance of Tregs depends on context and spatial localization. Tregs enriched within tumor nests or at key trafficking bottlenecks may have outsized impact compared with Tregs in peripheral stroma [21]. Some tumors also form tertiary lymphoid structures (TLS), which can support local antigen presentation and clonal expansion; however, TLS can also harbor regulatory circuits depending on cellular composition and cytokine balance. This spatial nuance helps explain why broad Treg depletion is not an attractive strategy in most settings, and why interventions that selectively modulate intratumoral regulation are of greater interest [22].

Contemporary approaches therefore aim for selective modulation of intratumoral Tregs or disruption of Treg recruitment. Examples include targeting receptors and pathways

preferentially expressed on tumor-resident Tregs, and interrupting chemokine axes that enrich Tregs within tumors [23]. Clinically, the goal is often to tilt the local balance toward effector dominance without provoking systemic autoimmunity. This makes combination design and dosing critical: modest shifts in the effector-to-regulatory ratio can be sufficient when paired with checkpoint blockade, whereas aggressive systemic modulation can raise toxicity without proportionate benefit [24].

The stromal fortress: breaking physical exclusion

Immune exclusion is a recurring phenotype in which cytotoxic lymphocytes accumulate at the invasive margin but fail to penetrate malignant nests. Cancer-associated fibroblasts (CAFs) and the extracellular matrix (ECM) they remodel are central contributors [25]. CAFs can deposit dense collagen networks that increase tissue stiffness, generate chemokine gradients that misdirect immune cells into non-productive compartments, and elevate interstitial fluid pressure that impairs immune cell motility and drug transport [26]. Beyond physical obstruction, CAFs can shape the immune milieu by secreting cytokines and growth factors that support suppressive myeloid states and by establishing niches that favor regulatory cells [27].

TGF-beta signaling frequently acts as a master regulator of exclusionary stroma. It reinforces fibroblast activation and ECM deposition and can directly suppress cytotoxic programs in T cells and natural killer cells [28]. Tumors with stromal TGF-beta activity are repeatedly associated with resistance to checkpoint blockade, supporting the concept that access barriers can dominate even when inhibitory receptors are targetable. Importantly, exclusion is often multifactorial: fibroblast programs intersect with abnormal vasculature, and both can raise pressure and alter chemokine gradients. Therefore, successful “breaching” of exclusion often requires addressing more than one stromal or transport feature [29].

Strategies to overcome exclusion aim to change the physical and chemical rules of the tissue. Approaches include targeting TGF-beta pathway activity, disrupting fibroblast-mediated matrix remodeling, and reducing stiffness or pressure that restricts cell movement. Because stromal barriers often sit upstream of inhibitory signaling, sequencing matters: enabling immune access may be a prerequisite for checkpoint blockade to act on a relevant population within tumor nests. Tissue-based spatial measurement is particularly valuable here, because it can quantify whether an intervention truly increases penetration into nests rather than simply increasing stromal accumulation at the border [30].

The priming bottleneck: reigniting antigen recognition

Tumors that appear as immune deserts often fail upstream at the level of antigen presentation and priming. Even if tumors harbor potential neo-antigens, inadequate recruitment and maturation of dendritic cells can limit cross-presentation and prevent expansion of tumor-reactive clones. Defects in innate sensing—whether tumor-intrinsic or microenvironmental—can further blunt priming, leaving checkpoint blockade without a meaningful substrate. In addition, some tumors present antigens but do so in a context dominated by suppressive cytokines or regulatory cells, leading to ineffective or tolerogenic priming rather than productive immunity [31, 32].

Priming strategies aim to generate productive inflammation and broaden antigen-specific repertoires. Radiotherapy can induce

immunogenic cell stress and antigen release [33]. Selected cytotoxic regimens can provide similar effects, though the balance between immunogenicity and lymphodepletion varies by drug and schedule [34]. Oncolytic platforms can provide infection-like cues that stimulate innate sensing and antigen presentation. Intratumoral delivery of innate agonists can activate antigen-presenting cells and promote chemokines that recruit effector cells. The objective is to couple antigen release with dendritic cell activation and trafficking to lymphoid sites so that primed T cells can expand and then access tumor tissue [35].

A practical challenge is that priming must be followed by effective trafficking and sustained effector function. When priming succeeds, inhibitory ligands and regulatory programs often rise as an adaptive response. This creates a predictable therapeutic sequence: initiate or amplify priming, then apply inhibitory pathway blockade and, when needed, suppression-relief modules. Without this stepwise logic, priming interventions can generate inflammation that fails to translate into tumor killing or that leads to transient responses followed by rapid adaptive escape [36].

Tumor-Intrinsic Immune Escape Programs

Cell-extrinsic suppression is only part of immune escape. Many tumors deploy intrinsic programs that reduce antigen visibility, blunt inflammatory signaling, or shape chemokine output in ways that favor exclusion and suppression. These mechanisms are clinically important because they can drive resistance even in the presence of immune infiltration and because they can emerge under therapy as a form of immune editing. Tumor-intrinsic escape is also a major reason why biomarkers based on a single biopsy can fail: the tumor compartment itself can evolve, and different regions can harbor distinct visibility and signaling states [37, 38].

Antigen presentation and immune visibility

Effective T-cell recognition requires intact antigen processing and presentation through major histocompatibility complex (MHC) class I. Tumors may downregulate this machinery through genetic alterations, epigenetic repression, or transcriptional adaptation. The result can be partial invisibility in which some clones remain visible while others become selectively advantaged under immune pressure. Such heterogeneity is common and can be missed by limited sampling, especially when immune-edited subclones occupy only a fraction of the tumor volume [39].

Loss or reduction of antigen presentation has practical implications for therapy selection and for expectations of checkpoint responsiveness. It can also inform combination logic. Strategies that enhance antigen presentation, promote re-expression of silenced components, or improve dendritic cell cross-presentation may increase the probability of sustained control. In some contexts, engaging immune effectors that are less dependent on MHC class I recognition, such as natural killer cells, may provide a complementary route. The broader lesson is that restoring immune recognition may require interventions directed at both the tumor and the microenvironment [40].

Interferon signaling and inflammatory competence

Interferon signaling can amplify anti-tumor immunity by enhancing antigen presentation, promoting chemokine gradients that recruit effector cells, and supporting cytotoxic effector programs [41]. Tumors with impaired interferon responsiveness may be less capable of sustaining inflammatory amplification and less vulnerable to immune-mediated killing. At

the same time, chronic interferon exposure can contribute to adaptive resistance by inducing inhibitory ligands and shaping selection pressures that favor immune-evasive clones. This dual role underscores why timing matters: early interferon competence may predict response, while chronic interferon-driven adaptation may contribute to relapse [42].

From a translational perspective, interferon competence can influence whether priming strategies translate into durable infiltration and whether checkpoint reinvigoration leads to sustained control. When interferon pathway disruption is suspected, on-treatment sampling and integrated pathway readouts can help distinguish failure of priming from failure of effector function. This distinction matters because the first scenario calls for improved antigen presentation and innate activation, while the second scenario calls for suppression relief and tissue conditioning to restore function [43].

Oncogenic signaling, chemokines, and immune architecture

Oncogenic programs influence immune escape by modulating chemokines, antigen presentation, and stromal interactions [44]. Some tumor states reduce recruitment of dendritic cells and effector lymphocytes while favoring suppressive myeloid infiltration. Others promote fibroblast activation, vascular dysfunction, or matrix remodeling, indirectly driving exclusion. These programs help explain why two tumors with similar checkpoint ligand expression can have very different immune architectures and different probabilities of responding to the same therapy [45].

Mechanistically, tumor-intrinsic signaling can alter the entry points for immunity by shaping which myeloid and lymphoid cells are recruited and where they localize. Reduced dendritic cell recruitment can blunt priming even in the presence of antigen, while altered chemokine output can trap T cells in stroma rather than in nests. Clinically, this supports the view that immune phenotypes can be driven from within the malignant compartment, and that successful remodeling may require addressing tumor-intrinsic signaling together with microenvironmental barriers [46].

Plasticity and state transitions under therapeutic pressure

Tumor cell plasticity provides an additional route to immune escape without requiring new mutations. Stress-induced transitions can change antigen expression, increase resistance to cytotoxic killing, and intensify paracrine signaling with stromal and myeloid compartments. These transitions are often enriched at invasive fronts and in hypoxic niches where selection pressures are high. Plasticity can therefore couple with metabolic and stromal barriers: hypoxic regions select for resistant states, and resistant states can in turn reinforce exclusion and suppression through altered cytokine and chemokine output [47, 48].

Recognizing plasticity as an escape route reinforces the need for longitudinal assessment. Static baseline sampling may underestimate the capacity for state change under therapy. In practical terms, this supports treatment strategies that combine reinvigoration of immunity with interventions that limit the emergence of resistant states, and it highlights the value of early on-treatment measurements that can detect whether the tumor ecosystem is moving toward immune control or toward adaptive escape [49]. An integrative framework linking immune escape patterns with dominant barriers and therapeutic modules is summarized in Figure 1, with key features and suggested outlined in Table 1.

Table 1. Mechanism-based immune escape patterns and corresponding therapeutic modules.

Immune Phenotype	Dominant Barrier	Key Biological Features	Therapeutic Module	Suggested Biomarker/Readout
Immune Desert	Priming failure	Low CD8, poor antigen presentation	Radiotherapy, oncolytic therapy, innate agonists	CD8 density, DC markers
Immune Excluded	Stromal barrier	TGF-β activation, fibroblast dominance	TGF-β blockade, stromal modulation	Spatial CD8 exclusion index
Inflamed but Suppressed	Functional exhaustion	PD-1/LAG-3 high, myeloid suppression	Dual checkpoint blockade, myeloid targeting	Exhaustion signature
Metabolic Niche	Hypoxia/adenosine	HIF-1α, CD39/CD73 activation	Adenosine pathway inhibitors	Hypoxia gene signature
Adaptive Escape	Antigen loss	MHC downregulation, clonal evolution	NK engagement, antigen restoration	MHC expression

Engineering the Metabolic and Physical Landscape

Even when effector cells breach stromal barriers, the metabolic and physical conditions of the TME can paralyze function. Uneven perfusion, hypoxia, acidosis, and nutrient competition erode immune cell fitness and can create localized suppressive niches. These constraints often act as invisible checkpoints that limit cytotoxicity even when inhibitory receptors are blocked. Because metabolic and physical barriers are frequently coupled to vascular and stromal architecture, they are attractive targets for tissue-conditioning strategies that can enable other immune-directed therapies [50].

Hypoxia and angiogenesis: normalizing supply lines

Tumor vasculature is typically disorganized and leaky, producing high interstitial pressure and uneven perfusion that oppose immune trafficking and drug delivery [17]. Poor perfusion generates hypoxic niches that stabilize hypoxia-responsive programs, promote suppressive myeloid states, and impair cytotoxic lymphocyte function. Hypoxia can also increase expression of inhibitory ligands, foster adenosine production, and exacerbate acidosis, linking vascular dysfunction to metabolic suppression [51].

Vascular normalization aims to remodel vessels so that perfusion improves and hypoxia decreases, rather than indiscriminately destroying vessels [52]. In principle, improved perfusion can increase immune cell entry, improve delivery of antibodies and cellular therapies, and shift the tissue toward a less suppressive state. Clinically, the benefit of vascular modulation likely depends on dose, timing, and the balance between improving access and avoiding excessive pruning that worsens hypoxia. When successful, vascular normalization can convert the vasculature from a barrier into a conduit for immune cells and therapeutics [53].

An important practical implication is that vascular interventions are enabling rather than purely cytotoxic. Their impact may be best captured by intermediate biological readouts such as improved perfusion, reduced hypoxia-associated programs, and increased intratumoral immune entry, rather than by direct tumor shrinkage alone. This aligns naturally with adaptive immunotherapy, in which early changes in tissue conditions inform whether additional modules should be

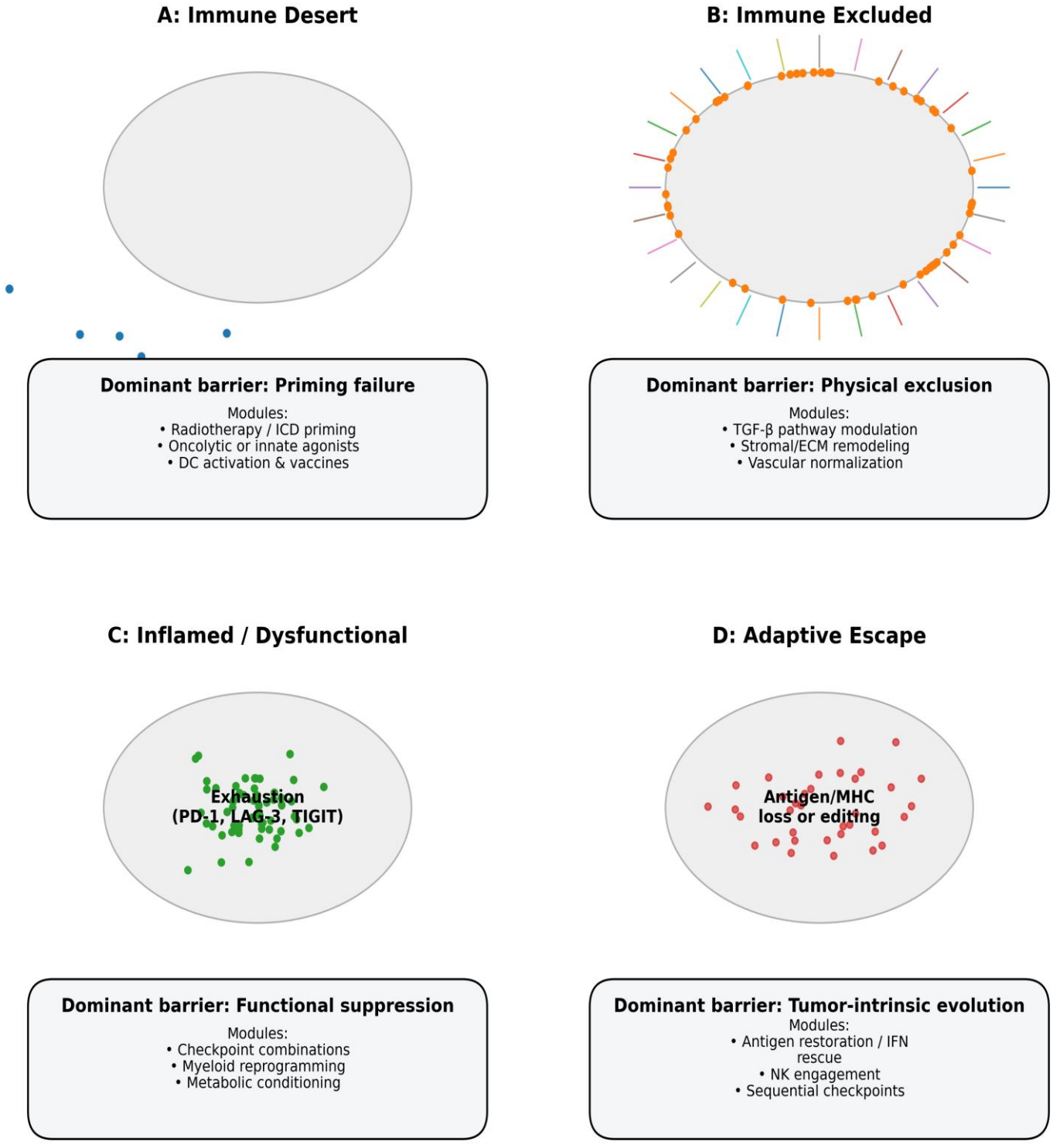

Figure 1. Mechanism-based classification of immune escape and therapeutic modules. *Four major immune escape phenotypes are summarized: immune desert (priming failure), immune excluded (stromal/physical barriers), inflamed but dysfunctional (checkpoint-driven exhaustion and suppressive conditioning), and adaptive escape (tumor-intrinsic evolution such as antigen/MHC loss). Representative mechanism-aligned therapeutic modules are listed for each phenotype.*

added or whether an alternative strategy is needed [54].

Metabolic checkpoints: adenosine, lactate, and pH

Aerobic glycolysis and impaired clearance of metabolites lead to lactate accumulation and extracellular acidification. Low pH can suppress T-cell cytokine production, impair lytic granule release, and favor suppressive polarization of innate cells [55]. In parallel, tissue stress and hypoxia can drive conversion of extracellular nucleotides into adenosine, which inhibits T-cell and natural killer cell activity through adenosine receptor signaling. Because ATP release can serve as an inflammatory danger signal, its conversion into adenosine is a particularly effective way for tissues to neutralize immune activation [56].

Targeting metabolic checkpoints is attractive but complicated by spatial heterogeneity. Metabolic suppression is rarely uniform; it often concentrates in micro-niches defined by perfusion, hypoxia, and stromal boundaries. This is one reason why spatially resolved measurements can matter: bulk signatures may miss dominant suppressive zones [57]. Therapeutically, interventions that block adenosine pathways or modulate lactate transport may be most effective when paired with strategies that increase effector presence, because relieving metabolic suppression has the most impact when functional immune cells are present to exploit improved conditions [58].

Metabolic interventions can influence multiple cell types beyond T cells. By shifting the constraints that favor suppressive macrophage states or that impair dendritic cell function, they may indirectly improve antigen presentation and reduce myeloid dominance. This broader impact supports combination designs in which metabolic checkpoint targeting is used as a tissue-conditioning module rather than as a stand-alone therapy [59].

Mechanical forces, ECM stiffness, and interstitial pressure

Beyond chemistry, the physical properties of tumors can constrain immunity [60]. Dense ECM networks, increased stiffness, and elevated interstitial pressure restrict immune cell movement and compartmentalize immune-tumor interactions. Mechanical cues also shape fibroblast and endothelial behavior, reinforcing exclusionary architectures and limiting efficient trafficking through vessels and stroma. The result can be an environment where immune cells are present but confined to areas in which they cannot exert cytotoxic effects [61].

Interventions that reduce matrix density or pressure may therefore serve as enabling steps that improve access and enhance the effectiveness of checkpoint blockade or other immune-directed therapies. From a practical standpoint, these approaches highlight that immune escape can be as much about tissue mechanics and transport as about receptor-ligand biology. They also emphasize the importance of integrating imaging and spatial pathology readouts into biomarker strategies, because mechanical barriers often manifest as structural patterns that can be quantified [62].

Checkpoint Ecosystems and T-Cell Resilience

In inflamed tumors, immunity is often present but functionally constrained. This dysfunction is maintained by a coordinated network of inhibitory receptors, suppressive ligands, and tissue signals that promote exhaustion-like programs. Understanding checkpoint biology therefore requires moving beyond a single marker and toward a view of inhibitory ecosystems and cellular states, including the surrounding metabolic and

myeloid context that determines whether reinvigoration is possible [63].

The exhaustion spectrum and reinvigoration potential

Chronic antigen exposure can push T cells into dysfunctional states characterized by reduced proliferation, diminished cytokine output, and impaired cytotoxicity. However, dysfunctional T cells are heterogeneous. Subsets with progenitor-like features can expand and regain function after inhibitory pathway blockade, whereas more terminally dysfunctional states may be less reversible. The distribution of these states can influence depth and durability of response, and it is shaped by antigen load, cytokine context, and tissue conditions such as hypoxia and nutrient availability [64].

Spatial localization is particularly important. Progenitor-like subsets often reside in specific niches, including regions near antigen-presenting cells or within TLS-like structures, and they may supply more differentiated effectors that enter tumor nests. If these supportive niches are absent or dominated by suppression, checkpoint blockade may have limited reinvigoration capacity even when PD-1 is expressed. This helps explain why PD-L1 staining alone is an incomplete predictor and why measuring both state and structure can improve mechanistic classification [65].

Compensatory inhibitory networks

Blocking one inhibitory pathway can unmask or induce others. Co-inhibitory receptors such as LAG-3, TIM-3, and TIGIT are often co-expressed with PD-1 on dysfunctional T cells and may contribute to incomplete or transient responses. These pathways are not interchangeable; they can operate through distinct ligands, cellular contexts, and downstream programs. A practical implication is that effective combinations should be selected based on the dominant inhibitory circuit within the relevant micro-niches, rather than applied as a generic escalation [66].

Compensatory inhibition also occurs at the level of ligands and accessory cells. Myeloid and stromal populations can upregulate inhibitory ligands under inflammatory pressure, and tumor cells can adaptively change their surface expression in response to therapy. Therefore, resistance after initial response may reflect a shift in the inhibitory ecosystem rather than the appearance of an entirely new escape mechanism. Early on-treatment measurement can guide rational escalation: if a compensatory pathway becomes dominant, targeting that pathway is more rational than adding agents unrelated to the evolving bottleneck [67].

Primary versus acquired resistance as a TIME problem

Primary resistance often reflects upstream barriers such as lack of priming, exclusionary stroma, myeloid dominance, or severe metabolic hostility. In these settings, checkpoint blockade is downstream of the limiting step and may have minimal effect. Acquired resistance more commonly reflects immune editing and adaptive remodeling under treatment pressure, including altered antigen presentation, disrupted inflammatory signaling, compensatory inhibitory receptor upregulation, and expansion of suppressive populations. This distinction matters because it frames the therapeutic task: initiating immunity, enabling access, restoring function, or countering adaptive escape [68].

Technology-Enabled Precision: Seeing the Invisible

Mechanistic insight becomes clinically useful only when it can be measured in a feasible and reproducible manner. Traditional single-marker assays and bulk averages often

miss spatial architecture, state heterogeneity, and dynamic change. Technology-enabled profiling is reshaping how the TIME is classified and how therapeutic strategies are selected. The most valuable approaches are those that preserve tissue context, allow standardization, and can be linked to decision frameworks that clinicians can apply without requiring high-dimensional data interpretation at the bedside [69].

Multiplex imaging and spatial contexture

Multiplex immunohistochemistry and immunofluorescence, as well as related imaging platforms, allow simultaneous evaluation of multiple markers while preserving tissue architecture. These methods can quantify not only cell densities but also critical spatial relationships, such as whether CD8+ T cells enter tumor nests or remain stromal, whether suppressive myeloid cells cluster around vessels, and whether fibroblast-rich borders form exclusionary fences. Spatial metrics support more faithful immune phenotyping because they describe the physical reality that determines whether immune cells can contact tumor cells [70]. A practical advantage is that multiplex imaging can often be deployed on standard formalin-fixed paraffin-embedded material. The main limitations relate to panel selection, analytic standardization, and sampling: a single biopsy may miss key niches, and different regions of the same tumor can show distinct architectures. These limitations argue for thoughtful sampling strategies and for reporting spatial outputs in ways that are interpretable and reproducible, such as quantifying border penetration, tumor-nest localization, and distances between effector and suppressive populations [71].

Spatial transcriptomics and proteomic mapping

Spatially resolved molecular profiling links gene or protein programs to micro-niches such as hypoxic cores, invasive margins, or tertiary lymphoid structures. These approaches can uncover compartment-specific suppression—such as stromal TGF-beta activity, hypoxia-adenosine programs, or localized myeloid polarization—that may be diluted in bulk assays. Their value increases when interpreted together with histology, connecting molecular signals to recognizable structures and enabling more precise hypotheses about dominant barriers [72].

As these platforms mature, their translational promise will depend on clarity of outputs. Clinicians need summaries that map to actionable choices rather than high-dimensional displays. One practical approach is to use spatial profiling to identify which barrier dominates which compartment, then link that to module selection: a tumor with a stromal TGF-beta border and poor intratumoral CD8 penetration suggests access modules, whereas a tumor with intratumoral CD8 cells co-localizing with adenosine and hypoxia programs suggests metabolic conditioning alongside checkpoint blockade [73].

Single-cell profiling to resolve functional states

Single-cell profiling refines the understanding of immune and stromal states, including gradients of T-cell dysfunction, dendritic cell maturation programs, and myeloid polarization states [74]. Although less scalable for routine practice, these approaches inform biomarker development by identifying cell states most strongly associated with response and resistance and by clarifying which pathways are plausibly causal rather than merely correlative. They also help define which cell populations express candidate targets,

which matters for anticipating on-target effects and for interpreting trial outcomes [75].

A recurring translational value of single-cell data is the identification of state markers that can be measured with simpler assays. If a progenitor-like exhausted T-cell subset is strongly linked to durable response, this can motivate focused immunohistochemistry panels or targeted gene expression readouts that approximate that state without requiring single-cell sequencing in every patient. Similarly, defining macrophage state programs can inform practical multiplex panels that distinguish suppressive from antigen-presenting compartments [76].

Digital pathology and quantitative image analysis

Quantitative analysis of routine pathology slides can provide scalable estimates of immune architecture, including tumor-stroma interfaces, immune clustering, and exclusion patterns. When validated across cohorts, these approaches can deliver low-friction phenotyping that complements more complex omics platforms. The most clinically useful outputs are those that remain interpretable, reproducible, and linked to a clear therapeutic logic, such as identifying exclusionary borders, myeloid-rich vascular niches, or patterns consistent with poor priming [77]. Quantitative image analysis should be viewed as a tool for standardization rather than as a substitute for mechanistic reasoning. Its value increases when it measures concepts clinicians already recognize—density, localization, border penetration, and structure—and when it is paired with biomarkers that capture functional state. This combination can move decision-making beyond subjective impressions and toward reproducible metrics suitable for trial stratification and, eventually, for clinical guidance [78].

Circulating biomarkers for longitudinal monitoring

Blood-based assays provide a complementary view that is well suited to longitudinal assessment. Dynamics of circulating tumor DNA can offer early indications of response or emerging resistance, while circulating immune analytes and cell populations may reflect systemic activation or suppression. Because circulating measures generally lack spatial information, they are best interpreted alongside tissue-based phenotyping: tissue defines the dominant local barrier, while serial blood measures track whether the tumor burden and systemic immune signals are moving in the expected direction after a given module is applied [79].

Longitudinal monitoring is particularly valuable for adaptive strategies. If a priming module is expected to increase immune engagement, one may look for early molecular signs of tumor stress and immune activation, followed by tissue confirmation when feasible. Similarly, rising circulating tumor DNA after an initial decline may signal emerging escape before radiographic progression, creating a window for rational escalation based on likely mechanisms rather than delayed clinical deterioration [80].

Integrating modalities into actionable phenotypes

The central translational challenge is to convert complex data into categories that guide decisions. A practical approach is to classify tumors into desert, excluded, or inflamed phenotypes using spatially informed tissue profiling, then refine that classification with pathway readouts that identify the dominant barrier (for example myeloid dominance, stromal TGF-beta activity, adenosine-rich niches, or compensatory inhibitory receptor networks). Early on-treatment reassessment can

further distinguish whether a therapy is shifting the intended barrier or merely changing peripheral markers without altering intratumoral biology [81].

Implementation considerations for clinical translation

For technology-enabled profiling to influence care, it must fit within real-world constraints: limited tissue, variable sampling sites, and the need for standardized reporting. Practical implementation therefore depends on selecting assays that are robust to pre-analytic variation, defining quality control steps, and reporting outputs that map to decisions. For example, reporting "CD8 density" alone is less informative than reporting CD8 localization in nests versus stroma, proximity to suppressive myeloid cells, and evidence of exclusionary borders. Similarly, pathway readouts should be presented as interpretable scores or categories (for example "high stromal TGF-beta activity" or "adenosine-enriched hypoxic niche pattern") that correspond to access or conditioning modules [82].

Another recurrent issue is heterogeneity across lesions and overtime. A primary tumor and metastasis can have different TIME architectures, and a tumor can transition from excluded to inflamed under therapy without becoming sensitive. This argues for longitudinal approaches when feasible, and for integrating tissue-based information with circulating monitoring. It also suggests that biomarker strategies should be explicit about which lesion is sampled and should avoid over-generalization from a single small biopsy. In clinical trials, adaptive designs and embedded translational endpoints can help establish which measurements best predict benefit and which modules actually shift the intended biology [83]. The main technology platforms used to map the tumor immune microenvironment and their potential for clinical translation are presented in Table 2.

Table 2. Technology-enabled platforms for tumor immune profiling and clinical integration.

Platform	Resolution	What It Measures	Clinical Value	Limitations
Multiplex IHC/IF	Spatial protein	CD8 localization, myeloid clusters	Phenotype classification	Limited gene depth
Spatial Transcriptomics	Spatial RNA	TGF-β niche, hypoxia	Mechanism mapping	Cost
Single-cell RNA-seq	Cellular state	Exhaustion spectrum	State-specific targeting	No spatial context
Digital Pathology	Quantitative morphology	Exclusion metrics	Standardization	Algorithm variability
Circulating Biomarkers	Systemic	ctDNA, immune activation	Dynamic monitoring	Low specificity

Mechanism-Guided Therapeutic Modules and Sequencing

Many tumors require multiple enabling steps before checkpoint blockade can produce durable control. A mechanism-guided strategy therefore assembles therapeutic modules that address the dominant barrier in each tumor and considers sequencing based on biology and feasibility. This approach aligns with the observation that different tumors fail for different reasons: some fail to generate tumor-reactive T cells, some prevent those cells from entering tumor nests, and some allow entry but maintain dysfunction through inhibitory and metabolic constraints. The objective is to match the intervention to the rate-limiting step, then reassess to determine what becomes limiting next.

Priming modules for immune-desert tumors

In desert tumors, the initial objective is to generate productive priming and recruitment. Modules include localized antigen release paired with inflammatory cues, intratumoral innate activation, and strategies that enhance dendritic cell recruitment and function. The goal is to expand the repertoire of tumor-reactive T cells and to create chemokine gradients that support trafficking into the tumor. Successful priming often leads to induction of inhibitory ligands and regulatory programs, creating a rationale for adding inhibitory pathway blockade once infiltration and antigen-specific responses are established [84].

A pragmatic concern is that priming can be ineffective if access barriers and metabolic constraints are not simultaneously addressed. Priming may increase circulating tumor-reactive T cells without changing intratumoral entry if vessels and stroma remain restrictive. Therefore, priming modules are often paired with access conditioning or scheduled in a sequence that first improves entry points and then applies strong priming. This logic is rarely captured by simplistic “add-on” combinations; it requires an explicit model of what is expected to change and when [85].

Access modules for immune-excluded tumors

In excluding tumors, access is the bottleneck. Modules include stromal remodeling strategies, TGF-beta pathway modulation, and vascular normalization approaches that improve trafficking and reduce hypoxia. Once access improves, inhibitory signaling and myeloid suppression often become the next limiting steps, supporting stepwise combinations that preserve effector function within tumor nests. Access modules are also relevant for cellular therapies and antibody-based platforms that require physical delivery into tumor tissue [36].

Access can be monitored using spatial metrics: increases in intratumoral CD8 density, reduced stromal trapping, and improved proximity between effectors and tumor cells. When access improves without tumor control, this suggests that dysfunction and suppression, rather than exclusion, are now dominant. This provides a concrete rationale for escalation toward checkpoint ecosystem targeting, myeloid reprogramming, or metabolic conditioning rather than for repeated attempts at further stromal disruption [1].

Function modules for inflamed but ineffective tumors

Inflamed tumors may fail because inhibitory circuits and suppressive networks keep effector cells below a functional threshold [86]. Modules include blockades of dominant checkpoint networks, myeloid reprogramming, and relief of metabolic suppression. The guiding principle is to match the intervention to

the most active suppressive circuit rather than applying maximal combinations indiscriminately, which can increase toxicity without addressing the true bottleneck. In practice, this may involve identifying compensatory inhibitory receptors, assessing myeloid dominance, and evaluating whether hypoxia-adenosine niches or low pH are likely to be paralyzing effector function [87].

Because inflamed tumors can still be heterogeneous, local micro-niches matter. A tumor can contain areas where T cells are active and areas where they are paralyzed by hypoxia or adenosine, leading to incomplete control and eventual escape. Function modules may therefore need to address both inhibitory receptors and tissue conditioning, especially in tumors with strong metabolic gradients. This is a setting where integrating spatial profiling with systemic monitoring can help distinguish true reinvigoration from superficial inflammatory changes [88].

Practical readouts for adaptive immunotherapy

A key opportunity is to couple module selection to early biological readouts. Examples include increases in intratumoral CD8 infiltration after access modules, shifts in myeloid state markers after reprogramming strategies, reduction of hypoxia-associated programs after vascular interventions, or emergence of compensatory inhibitory receptors after PD-1 blockade. Integrating such readouts with clinical endpoints can support adaptive strategies that escalate, sequence, or redirect therapy before overt progression. Importantly, the readout should match the intended mechanism: a priming module should be judged by evidence of new antigen-specific responses and recruitment, whereas an access module should be judged by spatial penetration rather than by peripheral cytokine changes [87, 89].

A pragmatic decision framework

A pragmatic framework is to start with phenotype-level classification and then confirm the dominant barrier with targeted measurements. Desert tumors prioritize priming; excluded tumors prioritize access; inflamed tumors prioritize restoration of function and suppression relief. In mixed phenotypes, the dominant barrier may be the one that prevents immune cells from reaching tumor nests or the one that suppresses the largest fraction of infiltrating effectors. This approach does not eliminate uncertainty, but it makes trial design and clinical decision-making more explicit: each intervention is paired with an expectation and a measurement plan, and failure becomes informative rather than ambiguous [7, 90, 91].

Conclusion and Future Perspectives

Immune escape is a system-level outcome of tumor-intrinsic programs, suppressive cellular networks, and hostile metabolic and biophysical constraints. This complexity explains why the same immunotherapy can be transformative in one patient and ineffective in another. Progress therefore depends on identifying the dominant barrier in each tumor and applying mechanism-aligned interventions that remodel the TIME.

Technology-enabled profiling is central to this shift because spatial organization and cellular state often determine whether infiltration translates into killing. Multiplex imaging, spatial profiling, single-cell discovery platforms, quantitative digital pathology, and longitudinal blood-based monitoring together provide a toolkit to classify tumors into actionable phenotypes and to track whether intended biological shifts occur under therapy.

Looking forward, the most informative clinical studies will be those that connect interventions to measurable biological changes. Adaptive trial designs that incorporate early on-treatment tissue or circulating assessments can test whether a proposed module truly shifts the expected barrier and whether that shift correlates with clinical benefit. In parallel, efforts to standardize spatial and state readouts will be essential so that results are comparable across centers and tumor types.

Ultimately, restoring durable anti-tumor immunity will likely require treating the TIME as a reprogrammable system: diagnose dominant barriers, apply targeted modules, and adapt as the ecosystem evolves. As measurement becomes more scalable and combination strategies become more mechanism-driven, TME engineering can move from an aspirational concept to a practical route for improving the depth and durability of immunotherapy responses across cancers.

Funding: *The author(s) received no financial support for the research, authorship, and/or publication of this article.*

Conflict of Interest: *The authors declare that they have no conflict of interest.*

Ethical statement: *It wasn't necessary.*

Author Contributions*: M.E. conceived the study, performed the literature review, and wrote the manuscript. The author approved the final version of the manuscript.*

References

[1]Chiodelli P, Papait A, Agoni L, Quaglia F, Silini AR, Parolini O. The tumor ecosystem: Rewiring an open, systemically integrated network for therapeutic gain. Biomed Pharmacother. 2026;195:119047.

[2]Rajbhandary S, Dhakal H, Shrestha S. Tumor immune microenvironment (TIME) to enhance antitumor immunity. Eur J Med Res. 2023;28(1):169.

[3]Torabi S, Yekzaman E, Taherkhani S, Talachi N, Khoshravesh R, Khosravi A, Salehi M, Zarrabi A, Vosough M. Immune cell-based therapies for solid tumors, current challenges and therapeutic advances. Cell Commun Signal. 2025;24(1):73.

[4]Abraham PE, Johnson DB. Long-Term Toxicities of Immune Checkpoint Inhibitors. Drugs. 2025;85(12):1535-1549.

[5]Kareva I, Gevertz JL. Mitigating non-genetic resistance to checkpoint inhibition based on multiple states of immune exhaustion. npj Systems Biology and Applications. 2024;10(1):14.

[6]Baldominos P, Barbera-Mourelle A, Barreiro O, et al. Quiescent cancer cells resist T cell attack by forming an immunosuppressive niche. Cell. 2022;185(10):1694-708. e19.

[7]Zheng S, Wang W, Shen L, et al. Tumor battlefield within inflamed, excluded or desert immune phenotypes: the mechanisms and strategies. Exper Hematol Oncol. 2024;13(1):80.

[8]Morello S, Pinto A, Blandizzi C, et al. Myeloid cells in the tumor

microenvironment: Role of adenosine. Oncoimmunol. 2016;5(3):e1108515.

[9] Huang Z, Chen J, Zhu T, Li J, Ng HY, Zhou Y, Gu X, Xu S, Jia R. Cancer-associated fibroblasts in the tumor microenvironment: heterogeneity, crosstalk mechanisms, and therapeutic implications. Mol Cancer. 2025;25(1):19.

[10] Jiang L, Qi Y, Yang L, et al. Remodeling the tumor immune microenvironment via siRNA therapy for precision cancer treatment. Asian J Pharm Sci. 2023;18(5):100852.

[11] Hanson GF, Goundry KA, Wessel RE, et al. A flexible systems analysis pipeline for elucidating spatial relationships in the tumor microenvironment linked with cellular phenotypes and patient-level features. Front Immunol. 2025;16:1642527.

[12] Ersoy M. Immature Granulocytes and Nivolumab Outcomes in Stage IV Non-Small Cell Lung Cancer: Insights by Tumor Subtype. Clinical Medicine Insights: Oncology. 2025;19:11795549251371094.

[13] Lee H, Kim B, Park J, et al. Cancer stem cells: landscape, challenges and emerging therapeutic innovations. Signal Transduct Target Ther. 2025;10(1):248.

[14] Yang Y, Li C, Liu T, et al. Myeloid-derived suppressor cells in tumors: from mechanisms to antigen specificity and microenvironmental regulation. Front Immunol. 2020;11:1371.

[15] Grzywa TM, Sosnowska A, Matryba P, et al. Myeloid cell-derived arginase in cancer immune response. Front Immunol. 2020;11:938.

[16] Chen H, Xu Z, Varner J. Targeting myeloid cells to improve cancer immune therapy. Front Immunol. 2025;16:1623436.

[17] Racacho KJ, Shiau Y-P, Villa R, et al. The tumor immune microenvironment: implications for cancer immunotherapy, treatment strategies, and monitoring approaches. Front in Immunol. 2025;16:1621812.

[18] Shi Z-D, Pang K, Wu Z-X, et al. Tumor cell plasticity in targeted therapy-induced resistance: mechanisms and new strategies. Signal Transduct Target Ther. 2023;8(1):113.

[19] Adam Essa ME, Elbadri S, Ahmed AA, et al. Reprogramming the tumour microenvironment: Emerging strategies to overcome immunotherapy resistance. Clin Trans Disc. 2025;5(6):e70098.

[20] Ge J, Yin X, Chen L. Regulatory T cells: masterminds of immune equilibrium and future therapeutic innovations. Front Immunol. 2024;15:1457189.

[21] Sato Y. The Role of Tregs in the Tumor Microenvironment. Biomedicines. 2025;13(5):1173.

[22] Baxevanis CN, Sofopoulos M, Tsitsilonis OE, et al. Exploring the Pivotal Functions of Tertiary Lymphoid Structures in Cancer Prognosis and Immunotherapy Outcomes. Cancers. 2025;17(23):3754.

[23] Mortezaee K. Selective targeting or reprogramming of intra-tumoral Tregs. Med Oncol. 2024;41(3):71.

[24] Cogdill AP, Andrews MC, Wargo JA. Hallmarks of response to immune checkpoint blockade. Br J Cancer. 2017;117(1):1-7.

[25] Bruni S, Mercogliano MF, Mauro FL, et al. Cancer immune exclusion: breaking the barricade for a successful immunotherapy. Front Oncol. 2023;13:1135456.

[26] Gamradt P, Thierry K, Masmoudi M, et al. Stiffness-induced cancer-associated fibroblasts are responsible for immunosuppression in a platelet-derived

growth factor ligand-dependent manner. Pnas Nexus. 2023;2(12):pgad405.

[27] Zhang F, Ma Y, Li D, et al. Cancer associated fibroblasts and metabolic reprogramming: unraveling the intricate crosstalk in tumor evolution. J Hematol Oncol. 2024;17(1):80.

[28] Ho MP, Jung M, Ung W, Skouradaki E, Baritaki S, Bonavida B. TGF-β-YY1 signaling as a key driver of immune evasion in pancreatic cancer: Therapeutic implications. Cytokine Growth Factor Rev. 2026;88:58-66.

[29] Hou W. Role of TGFβ-activated cancer-associated fibroblasts in the resistance to checkpoint blockade immunotherapy. Front Oncol. 2025;15:1602452.

[30] Frangogiannis NG. Transforming growth factor–β in tissue fibrosis. J Exper Med. 2020;217(3):e20190103.

[31] Bandola-Simon J, Roche PA. Dysfunction of antigen processing and presentation by dendritic cells in cancer. Mol Immunol. 2019;113:31-7.

[32] Xie Z, Fang Y, Zhang X, et al. The emerging role of dendritic cells in the tumor microenvironment: from antigen presentation to targeted immunotherapy. Cell Death Dis. 2025;16(1):900.

[33] Zhu S, Wang Y, Tang J, et al. Radiotherapy induced immunogenic cell death by remodeling tumor immune microenvironment. Front Immunol. 2022;13:1074477.

[34] Fabian KP, Wolfson B, Hodge JW. From immunogenic cell death to immunogenic modulation: select chemotherapy regimens induce a spectrum of immune-enhancing activities in the tumor microenvironment. Front Oncol. 2021;11:728018.

[35] Wang X, Wang J, Wang Q, et al. Precision cytokine modulation to overcome tumor microenvironment-driven resistance to immune checkpoint blockade. Biochim Biophysica Acta (BBA)-Rev Cancer. 2026:189536.

[36] Jiang S, Zhang Z, Mao Y, et al. Emerging nano-immunotherapeutic strategies achieve metastatic colorectal cancer precision therapy. J Nanobiotechnol. 2026.

[37] Ummarino A, Calà N, Allavena P. Extrinsic and cell-intrinsic stress in the immune tumor micro-environment. Inter J Mol Sci. 2024;25(22):12403.

[38] Sun J, Zamyatnin Jr AA, Yuan X, et al. Cancer cell-intrinsic and-extrinsic factors affecting tumor immune evasion. Front Med SA. 2023. p. 1261820.

[39] Shklovskaya E, Rizos H. MHC class I deficiency in solid tumors and therapeutic strategies to overcome it. Inter J Mol Sci. 2021;22(13):6741.

[40] O'Melia MJ, Menzel L, Lei PJ, et al. Overcoming impaired antigen presentation in tumor draining lymph nodes facilitates immunotherapy. bioRxiv [Preprint]. 2025 Sep 20:2025.09.17.676884.

[41] Ruiz-Iglesias A, Guilbaud E, Galluzzi L, et al. Context-dependent impact of type I interferon signaling in cancer. Mol Cancer. 2025;24(1):1-16.

[42] Massa C, Wang Y, Marr N, et al. Interferons and resistance mechanisms in tumors and pathogen-driven diseases—focus on the major histocompatibility complex (MHC) antigen processing pathway. Inter J Mol Sci. 2023;24(7):6736.

[43] Saleiro D, Platanias LC. Interferon signaling in cancer. Non-canonical pathways and control of intracellular immune checkpoints. Semin Immunol. 2019;43:101299.

[44] Tufail M, Jiang C-H, Li N. Immune evasion in cancer: mechanisms and cutting-edge

therapeutic approaches. Signal Transduct Targeted Ther. 2025;10(1):227.

[45] Yu J, Fu L, Wu R, et al. Immunocytes in the tumor microenvironment: recent updates and interconnections. Front Immunol. 2025;16:1517959.

[46] Ouyang P, Zhang J, He X, et al. Infiltration characteristics and regulatory mechanisms of CD8+ T lymphocytes in solid tumors: spatial distribution, biological functions, and interactions with the immune microenvironment. Front Immunol. 2025;16:1661545.

[47] Qin S, Jiang J, Lu Y, et al. Emerging role of tumor cell plasticity in modifying therapeutic response. Signal Transduct Targeted Ther. 2020;5(1):228.

[48] Gao D, Kan P, He Y, et al. Senescent immune cells in the tumor microenvironment: emerging insights into cancer immunotherapy resistance. Front Immunol. 2025;16:1656733.

[49] Ramisetty S, Subbalakshmi AR, Pareek S, et al. Leveraging cancer phenotypic plasticity for novel treatment strategies. J Clin Med. 2024;13(11):3337.

[50] Lim AR, Rathmell WK, Rathmell JC. The tumor microenvironment as a metabolic barrier to effector T cells and immunotherapy. Elife. 2020;9:e55185.

[51] Farhangnia P, Delbandi A-A, Aghamohammadi N, et al. The Role of Intra-Tumor Hypoxia in Cancer Cells Immune Escape Mechanism. Cancerous Cells: Immunobiology of Tumors and Metastasis: Springer; 2025. p. 297-345.

[52] Sun XX, Nosrati Z, Ko J, et al. Induced vascular normalization—can one force tumors to surrender to a better microenvironment? Pharmaceutics. 2023;15(8):2022.

[53] Mpekris F, Panagi M, Charalambous A, et al. Modulating cancer mechanopathology to restore vascular function and enhance immunotherapy. Cell Rep Med. 2024;5(7).

[54] Kim DJ. Molecular and Immune Mechanisms Governing Cancer Metastasis, Including Dormancy, Microenvironmental Niches, and Tumor-Specific Programs. Inter J Mol Sci. 2026;27(2):875.

[55] Gu X-Y, Yang J-L, Lai R, et al. Impact of lactate on immune cell function in the tumor microenvironment: mechanisms and therapeutic perspectives. Front Immunol. 2025;16:1563303.

[56] Audrito V, Messana VG, Brandimarte L, et al. The extracellular NADome modulates immune responses. Front Immunol. 2021;12:704779.

[57] Ferri-Borgogno S, Burks JK, Seeley EH, et al. Molecular, metabolic, and subcellular mapping of the tumor immune microenvironment via 3D targeted and non-targeted multiplex multi-omics analyses. Cancers. 2024;16(5):846.

[58] Zhang H, Li S, Liu S, et al. Therapeutic targeting of cell death-immune crosstalk in cancer to rewire the tumor immune microenvironment. Mol Cancer. 2025;24(1):1-36.

[59] Cote AL, Munger CJ, Ringel AE. Emerging insights into the impact of systemic metabolic changes on tumor-immune interactions. Cell Rep. 2025;44(2).

[60] Pourmostafa A, Uskach G, Jafari M, et al. Extracellular matrix physical properties regulate cancer cell morphological transitions in 3D hydrogel microtissues. Acta Biomater. 2026 Jan;210:17-26.

[61] Tiskratok W, Chuinsiri N, Limraksasin P, et al. Extracellular matrix stiffness: mechanotransduction and mechanobiological response-driven

strategies for biomedical applications targeting fibroblast inflammation. Polymers. 2025;17(6):822.

[62] Yang Y, Liu J, Liu J, et al. Advances in immune cell-based therapeutic agents for the treatment of inflammation-related diseases. Acta Pharmaceutica Sinica B. 2026. https://doi.org/10.1016/j.apsb.2026.01.013

[63] Chao Z, Mei Q, Yang C, et al. Immunological synapse: structures, molecular mechanisms and therapeutic implications in disease. Signal Transduct Targeted Ther. 2025;10(1):254.

[64] Huang Q, Li Y, Huang Y, et al. Advances in molecular pathology and therapy of non-small cell lung cancer. Signal Transduct Targeted Ther. 2025;10(1):186.

[65] Elfving H, Yu H, Fessehatsion KK, et al. Spatial distribution of tertiary lymphoid structures in the molecular and clinical context of non-small cell lung cancer. Cell Oncol. 2025;48(3):801-13.

[66] Wu Y, Wu Y, Gao Z, et al. Revitalizing T cells: breakthroughs and challenges in overcoming T cell exhaustion. Signal Transduct Targeted Ther. 2026;11(1):2.

[67] Sun Y, Shao C, Duan H, et al. Dynamic Evolution of the Tumor Immune Microenvironment in Malignant Tumors and Emerging Therapeutic Paradigms. MedComm. 2025;6(12):e70496.

[68] Alsaafeen BH, Ali BR, Elkord E. Resistance mechanisms to immune checkpoint inhibitors: updated insights. Mol Cancer. 2025;24(1):20.

[69] McKenzie M, Irac SE, Chen Z, et al., editors. Integrative Spatial Omics and Artificial Intelligence: Transforming Cancer Research with Omics Data and AI. Semin Cancer Biol. 2026: Elsevier.

[70] Lin J-R, Chen Y-A, Campton D, et al. High-plex immunofluorescence imaging and traditional histology of the same tissue section for discovering image-based biomarkers. Nature Cancer. 2023;4(7):1036-52.

[71] Mi H, Sivagnanam S, Ho WJ, et al. Computational methods and biomarker discovery strategies for spatial proteomics: a review in immuno-oncology. Brief Bioinform. 2024;25(5):bbae421.

[72] Wang Z-B, Long M-H, Yu P, et al. Spatial profiling of the metabolism-immune axis in ovarian cancer. Front Pharmacol. 2026;16:1672020.

[73] Khoshfekr Rudsari H, Tseng B, Zhu H, et al. Digital twins in healthcare: a comprehensive review and future directions. Front Digit Health. 2025;7:1633539.

[74] Marteau V, Nemati N, Handler K, et al. Single-cell integration and multi-modal profiling reveals phenotypes and spatial organization of neutrophils in colorectal cancer. Cancer Cell. 2026;44(1):146-65. e14.

[75] Prummel KD, Woods K, Kholmatov M, et al. Inflammatory stromal and T cells mediate human bone marrow niche remodeling in clonal hematopoiesis and myelodysplasia. Nature Commun. 2025;16(1):10042.

[76] Corselli M, Saksena S, Nakamoto M, et al. Single cell multiomic analysis of T cell exhaustion in vitro. Cytometry Part A. 2022;101(1):27-44.

[77] Willis J, Anders RA, Torigoe T, et al. Multi-institutional evaluation of pathologists' assessment compared to immunoscore. Cancers. 2023;15(16):4045.

[78] Lara H, Li Z, Abels E, et al. Quantitative Image Analysis for Tissue Biomarker Use: A White Paper From the Digital Pathology Association. Appl Immunohistochem Mol Morphol. 2021;29(7):479-493.

[79] Kaczmarek F, Marcinkowska-Gapińska A, Bartkowiak-Wieczorek J, et al. Blood-based biomarkers as predictive and prognostic factors in immunotherapy-treated patients with solid tumors—currents and perspectives. Cancers. 2025;17(12):2001.

[80] He M, Ji C, Li Z, et al. Circulating tumor DNA predicts clinical benefits of immune checkpoint blockade in HER2-negative patients with advanced gastric cancer: M. He et al. Gastric Cancer. 2025;28(5):872-85.

[81] Lodi F, Vanmassenhove S, Chen D, et al. Decoding tumor heterogeneity: A spatially informed pan-cancer analysis of the tumor microenvironment. Cell Reports Medicine. 2025;6(10).

[82] Cheng CH, Shi S-s. Artificial intelligence in cancer: applications, challenges, and future perspectives. Mol Cancer. 2025;24(1):274.

[83] Li M, Wang T, Zhang Z, et al. Metabolic reprogramming in the post-metastatic tumor microenvironment: multi-omics insights into determinants of immunotherapy response. Frontiers in Immunology. 2025;16:1742855.

[84] Xiao Z, Wang J, He S, et al. Engineered T cells stimulate dendritic cell recruitment and antigen spreading for potent anti-tumor immunity. Cell Rep Med. 2025;6(9).

[85] Luri-Rey C, Teijeira Á, Wculek SK, et al. Cross-priming in cancer immunology and immunotherapy. Nature Rev Cancer. 2025;25(4):249-73.

[86] Lems CM, Burger GA, Beltman JB. Tumor-mediated immunosuppression and cytokine spreading affects the relation between EMT and PD-L1 status. Front Immunol. 2023;14:1219669.

[87] Ma C, Li Y, Li M, et al. Targeting immune checkpoints on myeloid cells: current status and future directions. Cancer Immunol Immunother. 2025;74(2):40.

[88] Kunachowicz D, Tomecka P, Sędzik M, et al. Influence of hypoxia on tumor heterogeneity, DNA repair, and cancer therapy: from molecular insights to therapeutic strategies. Cells. 2025;14(14):1057.

[89] Reina-Campos M, Scharping NE, Goldrath AW. CD8+ T cell metabolism in infection and cancer. Nature Rev Immunol. 2021;21(11):718-38.

[90] Morain SR, Semler MW, Casey JD. A framework for health systems to assess and prioritize participation in pragmatic clinical trials. Learn Health Syst. 2025;9(4):e70016.

[91] Angeli S, Neophytou C, Charalambous A, et al. Modulating tumor mechanics and vascular barriers to enhance chemotherapy efficacy in pancreatic tumors and melanoma. J Control Release. 2025:114546.