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The interplay of effector and regulatory T cells in cancer

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Regulatory T (T_{reg}) cells suppress effector T (T_{eff}) cells and prevent immune-mediated rejection of cancer. Much less appreciated are mechanisms by which T_{eff} cells antagonize T_{reg} cells. Herein, we consider how complex reciprocal interactions between T_{eff} and T_{reg} cells shape their population dynamics within tumors. Under states of tolerance, including during tumor escape, suppressed T_{eff} cells support T_{reg} cell populations through antigen-dependent provision of interleukin (IL)-2. During immune activation, T_{eff} cells can lose this supportive capacity and directly antagonize T_{reg} cell populations to neutralize their immunosuppressive function. While this latter state is rarely achieved spontaneously within tumors, we propose that therapeutic induction of immune activation has the potential to stably disrupt immunosuppressive population states resulting in durable cancer regression.

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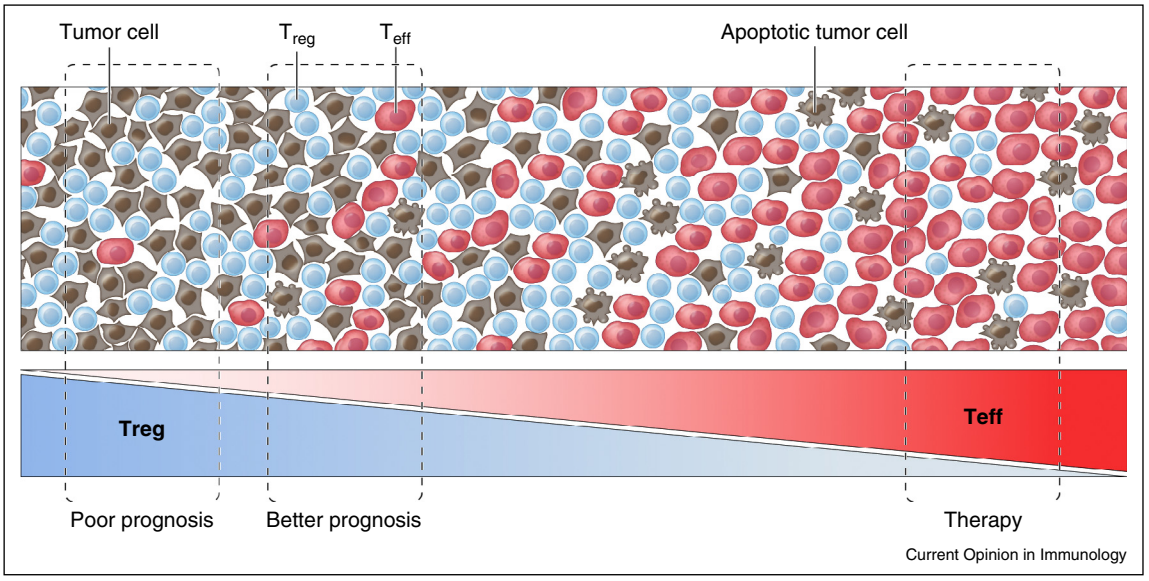
Introduction

Growth of tumors in immunocompetent hosts is at odds with the powerful ability of the immune system to recognize and kill cancer cells. The cancer immunoediting hypothesis has been proposed as a conceptual framework to account for this behavior [1]. According to this hypothesis, tumor development is characterized by an initial 'elimination' phase, during which a majority of cancer cells are destroyed by various components of the immune system. This is followed by an 'equilibrium' phase, during which pressure from the immune system contributes to selection of tumor variants that give rise to an 'escape' phase characterized by evasion from immune control and unrestrained tumor growth. While selection of antigen-loss variants represents a mechanism of tumor escape and has been shown to contribute to growth of orthotopic tumors [1–2], it fails to explain why established tumors continue to express immunogenic epitopes that are recognized by tumor-infiltrating lymphocytes and the efficacy of certain immune-based therapies for cancer [3–6,7••,8]. Growth of tumors containing immunogenic epitopes is better explained through an understanding of the critical role of immunosuppression in promoting tumor escape [9–12]. Here, we review recent advances in our understanding of tumor immunosuppression and consider how a complex interplay between T_{reg} and T_{eff} cell populations dictates the outcome of tumor-specific immune responses.

A major advance in our understanding of peripheral tolerance arose with the identification of a suppressive subset of $CD4^+$ T cells, referred to as T_{reg} cells, that express the high-affinity receptor for interleukin (IL)-2, IL-2Rα, and whose deficiency in neonatally thymectomized mice results in lethal inflammatory disease [13]. The similar inflammatory phenotype manifested in 'Scurfy' mice [14] was later attributed to a complete defect in T_{reg} cell formation caused by an inactivating mutation within the gene encoding the transcription factor (TF) Forkhead box P3 (Foxp3). This resulted in identification of Foxp3 as a lineage specifying TF of T_{reg} cells [15–17]. A broader network of TFs, including BACH2 [18••] and Foxo1 [19••,20,21], are required establish the full T_{reg} cell transcriptional program. Humans lacking a functional *Foxp3* locus develop a lethal immune-mediated disease (immunodysregulation polyendocrinopathy enteropathy X-linked syndrome; IPEX) [22], while genetic polymorphisms within the *BACH2* locus are associated with multiple autoimmune and allergic diseases [18••]. These findings provide evidence that T_{reg} cells regulate immune function in humans.

A complex interplay between $CD4^+$ T_{reg} and T_{eff} cells determines the outcome of immune reactions. Under homeostatic conditions, T_{reg} cells promote peripheral tolerance primarily through direct or indirect suppression of $CD4^+$ $Foxp3^-$ T_{eff} cells. This is evidenced by the absence of inflammation in *Cd4*-deficient mice which lack both T_{reg} and $CD4^+$ T_{eff} cell populations [23]. T_{reg} cells also suppress T_{eff} cell function within tumors (Figure 1). In murine tumor models, transient ablation of T_{reg} cells results in activation of $CD4^+$ or $CD8^+$ T_{eff} cells and rejection of solid tumors [24,25,26••,27]. In human tumors, low T_{reg} cell to T_{eff} cell ratios are associated with favorable survival in ovarian cancer [28,29], breast cancer [30], non-small cell lung carcinoma [31], hepatocellular carcinoma [32], renal cell carcinoma [33], pancreatic cancer [34], gastric cancer [35], cervical cancer [36]

Figure 1



The balance of T_{eff} and T_{reg} cells in tumors determines the functional outcome of immune responses. T_{reg} cells are represented in blue; T_{eff} cells are represented in red; tumor cells are depicted in brown. A spectrum of T_{eff}:T_{reg} cell ratios is depicted in the diagram going from left to right. Increased apoptosis is observed as the T_{eff}:T_{reg} cell ratio increases from left to right. The two boxes to the left represent the range of T_{eff}:T_{reg} cell ratios normally found within solid tumors. Within this range, the higher T_{eff}:T_{reg} cell ratios is associated with good prognosis in multiple cancer types (Refs. [23–25,26••,27–32]). The desired outcome of therapeutic manipulation of T_{eff}:T_{reg} cell ratios is depicted in the box to the right, where predominant T_{eff} cell populations mediate widespread tumor cell destruction and eradication of disease.

and colorectal carcinoma [37]. $CD4^+$ and $CD8^+$ T_{eff} cells exert tumoricidal activity through multiple means that are reviewed extensively elsewhere [38,39]. Thus, the balance between T_{reg} and T_{eff} cells dictates the outcome of tumor-specific immune responses (Figure 1). It is therefore important to understand factors that affect the population dynamics of T_{reg} and T_{eff} cells within tumors.

Population dynamics of T_{reg} and T_{eff} cells within tumors

The immunosuppressive function of T_{reg} cells is at odds with the capacity of the immune system to mediate brisk and functional T_{eff} cell responses against harmful pathogens in the context of acute infection. This implies that T_{reg} cell suppressive capacity may be neutralized during infection to promote clearance of disease. Emerging evidence presents multiple mechanisms by which T_{eff} cells antagonize the size and function of T_{reg} cell populations not only during infection, but also during inflammation and under specific conditions within tumors. Thus, while suppression of T_{eff} cells by a functionally predominant T_{reg} cell population represents the *status quo* during the 'escape' phase of tumor development, we propose that under specific conditions that either occur spontaneously or are induced therapeutically, a second state involving T_{eff}-mediated antagonism of T_{reg} cell populations can be induced that results in predominant T_{eff} cell function and clearance of disease.

The role of IL-2 signaling in T_{reg} and T_{eff} cell population dynamics

IL-2 was originally characterized as a lymphocyte growth factor *in vitro* and was considered to have immunostimulatory function [40,41]. Subsequent characterization using gene-deficient mice led to the surprising finding that its non-redundant biological function *in vivo* is to restrain lethal inflammation [42,43]. This is attributable to a non-redundant requirement for IL-2 in T_{reg} cell survival [44–46]. IL-2-driven induction of the anti-apoptotic Bcl-2 family member Mcl-1 is critical for IL-2-dependent survival of T_{reg} cells [47••].

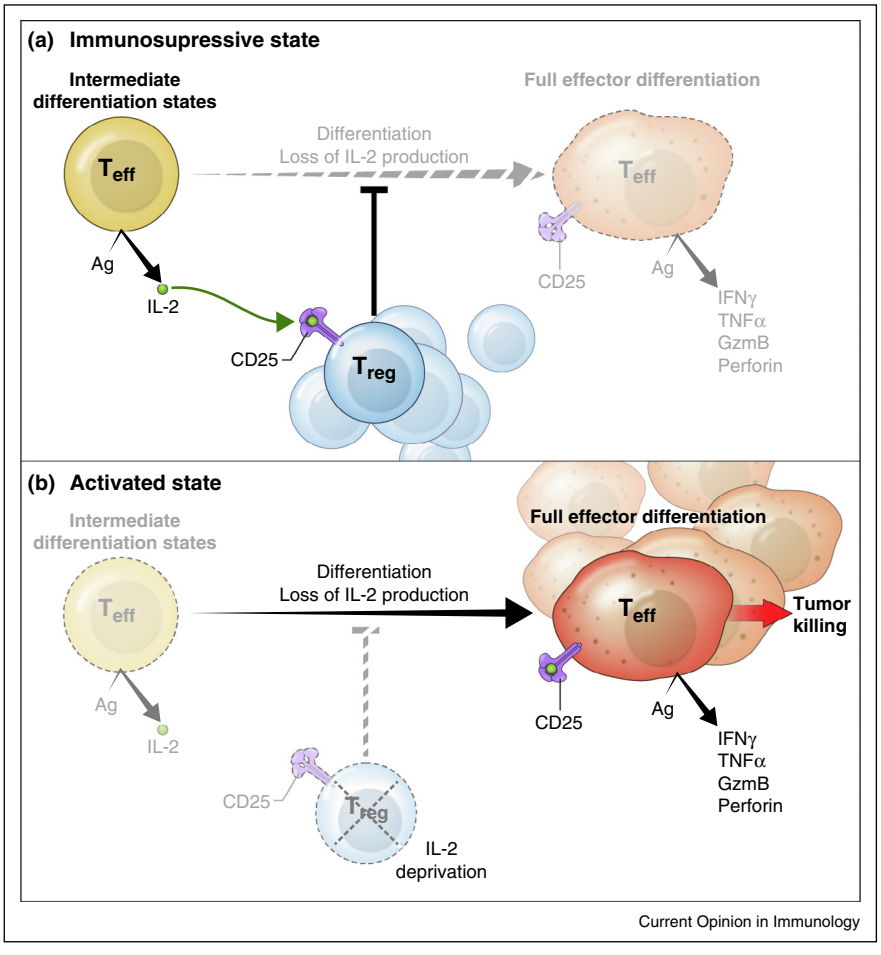
Suppressed T_{eff} cells support T_{reg} cells through paracrine IL-2 production

IL-2 is primarily produced by $CD4^+$ T cells and $CD8^+$ T cells and, to a lesser extent, NK cells. Since $Foxp3^+$ T_{reg} cells do not produce the cytokine, paracrine IL-2 generated in an antigen-dependent fashion by T_{eff} cells is required for T_{reg} cell survival [46,48]. This enables coupling of T_{reg} cell population size to the level of activation of T_{eff} cells at distinct sites providing a negative feedback loop that prevents excessive immune activation under homeostatic conditions [47••,49,50••]. However, not all T_{eff} cell populations produce IL-2 upon encounter with antigen and only ~10% of $CD4^+$ T cells are primed for IL-2 production upon antigen stimulation [50••]. In multiple T cell lineages, IL-2 expression is only associated

with activation of cells in an intermediate stage of differentiation and this is extinguished upon full effector differentiation. This process has been well characterized within the $CD8^+$ T cell lineage [51–54] and, to a lesser extent, within $CD4^+$ T cells [55,56]. Multiple signals, including IL-2 itself, contribute to loss of IL-2 production by T_{eff} cell populations [50••]. In part, this occurs through induction of the transcription factor PR domain zinc finger protein 1 (Blimp-1), that promotes acquisition of effector cell characteristics and loss of memory cell features including IL-2 production [56–60]. Thus, it is likely that T_{eff} cells in intermediate stages of differentiation, rather than terminally differentiated effector cells, predominantly contribute to T_{reg} cell maintenance [61••].

It is possible that T_{reg} cells actively maintain T_{eff} cells in intermediate stages of differentiation by suppressing full effector differentiation, resulting in stabilization of IL-2-producing populations of T_{eff} cells. T_{reg} cells play a critical role in formation of memory $CD8^+$ T cells by withholding IL-2 and preventing full effector differentiation in a subset of cells during the acute phase of primary $CD8^+$ T cell responses [62••]. Moreover, production of TGF-β by T_{reg} cells prevents full cytotoxic effector differentiation in tumor-specific $CD8^+$ T cells [63]. It is therefore plausible that T_{reg} cells, through sequestration of IL-2 and production of TGF-β, suppress full effector differentiation in T_{eff} cell populations to maintain a population of supportive IL-2-producing cells. This provides a potential feed-forward mechanism by which T_{reg} cell populations are supported by suppressed T_{eff} cells, constrained in intermediate stages of differentiation, to reinforce and stabilize the immunosuppressive state within tumors (Figure 2a).

Figure 2



IL-2: The currency of T_{reg} and T_{eff} cell population dynamics. **(a)** Immunosuppressive state. T_{reg}-mediated suppression of T_{eff} cells results in blockade of full effector differentiation. This maintains T_{eff} cells in intermediate states of differentiation that support T_{reg} cells through antigen-dependent paracrine IL-2 production. **(b)** Activated immune state. Reduced T_{reg} cell suppression results in full T_{eff} cell differentiation. This is accompanied by loss of antigen-dependent IL-2 production and withdrawal of IL-2 support for T_{reg} cells. Additionally, expression of CD25 by activated T_{eff} cells enables them to sequester any remaining IL-2.

Unrestrained T_{eff} cell differentiation results in withdrawal of paracrine IL-2 support

T_{eff} cells progressively loose the capacity to produce IL-2 upon effector differentiation [50••,51–53,55–60]. This raises the possibility that under certain conditions, unrestrained differentiation of T_{eff} cells results in withdrawal of cytokine support for T_{reg} cells. Such 'withdrawal' of cytokine support by T_{eff} cell populations is powerfully evidenced by the work of Oldenhove *et al.* in their study of T_{reg} and T_{eff} cell population dynamics during lethal infection with *Toxoplasma gondii* in mice [64]. Following infection, a striking collapse in T_{reg} cell frequency and absolute number coincides with loss of IL-2 production by Foxp3$^-$ CD4$^+$ T cells in the gut. This correlates with acquisition of a type helper 1 (Th1)-polarized state in which cells progressively lose expression of IL-2 and gain expression of IFN-γ as infection progresses toward its lethal outcome. Strikingly, complementation of IL-2 signaling through provision of exogenous IL-2 restores T_{reg} cell numbers and prevents lethality. These findings have been recapitulated in *Listeria monocytogenes* and vaccinia virus infection [65••]. Thus, induction of specific T_{eff} cell polarization states results in withdrawal of IL-2 support for T_{reg} cells, collapse in their population size, and unrestrained T_{eff} cell activation. Moreover, expression of IL-2Rα on T_{eff} cells is induced by antigen activation and terminal differentiation and may enable T_{eff} cells to sequester any remaining IL-2, further limiting its availability to T_{reg} cells [66,67].

Thus, complex regulation of IL-2 and its high-affinity receptor IL-2Rα results in the potential for bistability in T_{reg} and T_{eff} cell population dynamics within tumors. First, an immunosuppressive state exists where T_{reg} cells are supported by IL-2-producing T_{eff} cells constrained in intermediate states of differentiation (Figure 2a). This immunosuppressive state predominates during the escape phase of tumor development. Second, an activated immune state exists, in which unrestrained T_{eff} cell differentiation results in withdrawal of paracrine IL-2 support and competition for remaining IL-2 through antigen-driven expression of IL-2Rα (Figure 2b). While this state is rarely achieved spontaneously following tumor escape, such conditions may be achieved following therapeutic intervention (discussed below). The potential for IL-2 competition to account for population bistability is supported by mathematical models [68–70].

Reciprocal antagonism between T_{reg} and T_{eff} cell populations

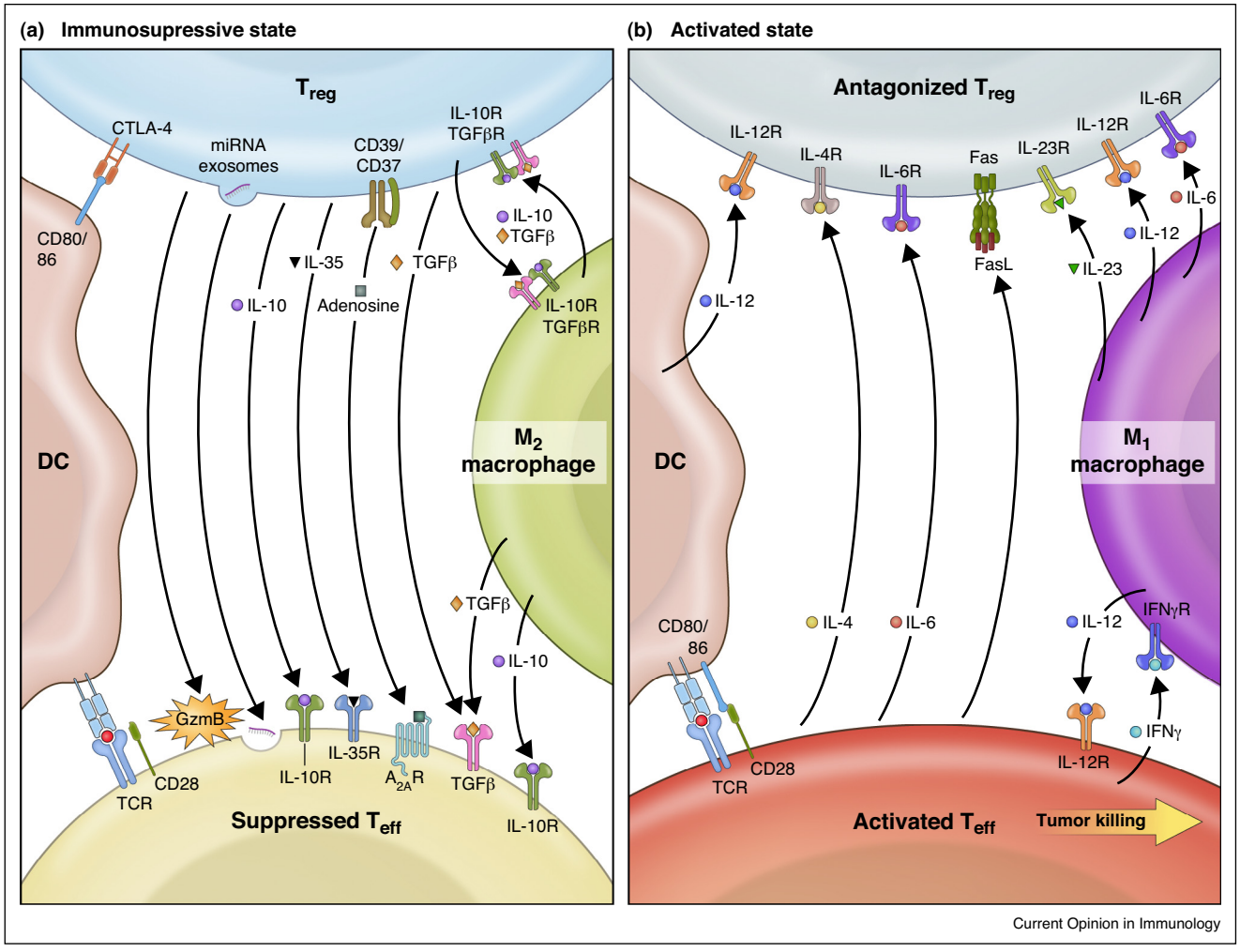
T_{reg}-mediated suppression of T_{eff} cell populations

In states of tolerance, including during tumor escape, T_{reg} cells block the proliferation, survival and function of T_{eff} cells through multiple means. These have been extensively reviewed [71] and are depicted in Figure 3a. A subset of Foxp3$^+$ T_{reg} cells constitutively express IL-2Rα and sequestration of IL-2 by T_{reg} cells is a component of their suppressive function [72,73]. However, expansion of T_{eff} cells and induction of lethal inflammation caused by T_{reg} cell insufficiency can proceed in the complete absence of IL-2 since IL-2 deficient mice succumb to lethal autoimmunity related to defective T_{reg} cell homeostasis [42,46,66], indicating redundant modes of suppression. These have been extensively reviewed elsewhere [71] and include CTLA-4-mediated sequestration of CD80 and CD86 on the surface of APC by trogocytosis [74] and expression of inhibitory cytokines such as TGF-β [75–77], IL-10 [78,79] and IL-35 [80,81]. Further, paracrine production of adenosine by T_{reg} cells suppresses T_{eff} cell differentiation and function [82,83]. Direct cytolysis of T_{eff} cells by T_{reg} cells is an additional mechanism of immunosuppression and has been attributed to expression of granzyme B and perforin by T_{reg} cells [84,85]. This was found to contribute to T_{reg}-mediated immunosuppression within tumors [86]. Finally, delivery of microRNA-containing exosomes is another mechanism of T_{reg}-mediated immunosuppression [87]. Innate immune cells contribute to feed-forward reinforcement of immunosuppressive states induced by T_{reg} cells. Amplification of inhibitory cytokine signaling by pro-tumorigenic tumor-associated macrophages (TAM) bearing an M2 (or alternatively activated) phenotype [88,89], and dendritic cells [9] are all implicated in promoting stability of immunosuppressive states within tumors.

Antagonism of T_{reg} cell populations by T_{eff} cells

During infection or upon induction of immune-mediated tumor rejection, emerging evidence indicates that T_{eff} cells may directly antagonize the stability and survival of T_{reg} cell populations to decrease their suppressive capacity, as depicted in Figure 3b. Selective killing of T_{reg} cells by T_{eff} cells is an example of such behavior. A decrease in T_{reg} cell numbers within tumors accompanies intratumoral IL-12 injection and this is dependent upon intrinstic expression of the death-receptor ligand FasL by CD8$^+$ T cells [90]. This observation is attributed to direct FasL-mediated killing of T_{reg} cells by T_{eff} cells. Induction of T_{reg} cell lineage instability is another mechanism by which immune activation within tumors may antagonize T_{reg} cell population size. The topic of T_{reg} cell lineage instability is controversial. By indelibly labeling cells and their progeny that had transcriptionally activated the endogenous *Foxp3* locus, Rubtsov *et al.*, concluded that T_{reg} cells rarely convert into Foxp3$^-$ 'ex-Foxp3' cells under physiological conditions or when perturbed during inflammation or lymphopenia [91]. However, using a similar approach based on a *Foxp3* reporter construct encoded by a bacterial artificial chromosome, Zhou *et al.*, observed accumulation of 'ex-Foxp3' IL-17-expressing cells in inflamed joints in response to synovial IL-6 [92]. Conversion of purified populations of Foxp3$^+$ T_{reg} cells into Foxp3$^-$ cells has also been observed in adoptive transfer models, both under conditions of extreme inflammation induced through allogeneic bone-marrow

Figure 3



Mechanisms of reciprocal antagonism between T_{reg} and T_{eff} cell populations. **(a)** Immunosuppressive state. T_{reg} cells suppress T_{eff} cell differentiation by multiple means including CTLA-4-dependent trogocytosis of CD80/86 on dendritic cells (DC), Granzyme B (GzmB) mediated killing of T_{eff} cells, production of inhibitory cytokines and small molecules (adenosine, IL-10, TGF-β and miRNA exosomes). Additionally, macrophages and dendritic cells provide feed-forward reinforcement of immunosuppressive states. **(b)** Activated immune state. Activated T_{eff} cells antagonize T_{reg} cells under conditions of immune activation. T_{eff} cells produce IL-6 and IFN-γ which leads to functional plasticity of T_{reg} cells. T_{eff} cell-derived IFN-γ causes dendritic cells and macrophages to produce IL-12 which results in a feed-forward cycle of IFN-γ production and induction of functional plasticity in T_{reg} cells. Additionally, exposure to inflammatory cytokines may stimulate T_{reg} cells to loose Foxp3 expression and acquire effector cell characteristics. Unrestrained differentiation and function of T_{eff} cell populations leads to immune activation and tumor killing.

transplantation [93], and extensive lymphopenia-induced proliferation [94]. These studies implicate the inflammatory cytokines IL-6 and/or IL-4, that are produced by T_{eff} cells, in driving lineage instability. It is therefore possible that under the inflammatory conditions induced by specific immune-based therapies for cancer, T_{reg} cell lineage instability contributes to reversal of tumor immunosuppression.

The balance of T_{reg} to T_{eff} cells is also modulated at the level of their induction. A proportion of T_{reg} cells found in peripheral tissues arise in the thymus (thymic T_{reg} or tT_{reg} cells) and play a predominant role in promoting tumor immunosuppression [95]. However, induced T_{reg} (iT_{reg}) cells develop from conventional Foxp3$^-$ naïve CD4$^+$ T cells in extrathymic tissues [96]. *De novo* induction of iT_{reg} cells is observed within tumors [97] and their suppressive function has been demonstrated in the context of therapeutic vaccination [98] and adoptive transfer immunotherapy [99]. A number of effector cytokines, including IL-4, IL-6 and IL-23 are involved in driving *de novo* induction of T_{eff} cell lineages from naïve CD4$^+$ T cell precursors [100] and this process is implicitly reciprocal to *de novo* induction of iT_{reg} cells from the same

precursor pool. Moreover, the T_{eff} cell cytokines IL-4, IFN-γ and IL-23 directly antagonize TGF-β-driven iT_{reg} cell induction [101–103].

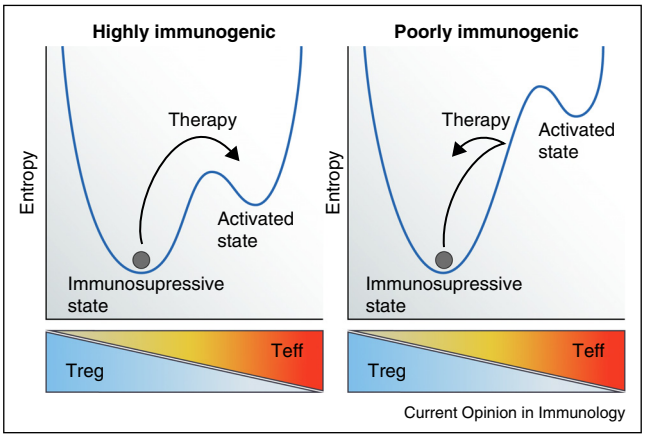
Finally, modulation of Foxp3$^+$ T_{reg} cell function is a distinct mechanism by which T_{eff} cells may alter the functional output of T_{reg} cell populations within tumors. Under conditions of lethal infection, Foxp3$^+$ cells expressing T-bet and IFN-γ are induced as a result of high levels of IL-12 secretion by DC within the lamina propria [64]. Similar plasticity is observed during viral infection of the central nervous system in mice [104]. Inflammatory conditions can also directly antagonize the suppressive function of T_{reg} cells. Using a transfer colitis model, the inflammatory cytokine IL-23 was found to promote colitis by inhibiting the suppressive function of Foxp3$^+$ T_{reg} cells [103].

Thus, direct antagonism of T_{reg} cell number and function equip T_{eff} cells with the capacity to disrupt T_{reg}-mediated immunosuppression once a specific threshold of activation has been achieved. Feed-forward reinforcement by innate immune cells within tumors contributes to the stability of activated immune states. In particular, Th1 cytokine-induced polarization of TAM into M1 (or classically activated) cells further potentiates immune activation through production of IL-12, IL-1, IL-6, TNF-α and IL-23 [88,89]. Additionally, dendritic cells provide further potential for feed-forward reinforcement by producing IL-12 in response to T_{eff} cell-derived IFN-γ.

Opportunities for therapeutic intervention

Reciprocal antagonism and feed-forward reinforcement contribute to the potential for two distinct immune states. First, an immunosuppressive state, which is established early during the escape phase of tumor development, is stabilized through provision of IL-2 support by suppressed T_{eff} cells for T_{reg} cells. Second, an activated immune state, in which unrestrained T_{eff} cell differentiation is accompanied by withdrawal of cytokine support and direct antagonism of T_{reg} cell populations to drive clearance of disease. Despite the potential availability of activated immune states, immunosuppressive states represent the *status quo* of tumor escape and entry into activated immune states is rarely achieved. However, under certain circumstances that either arise spontaneously [105,106] or in the context of therapeutic manipulation, transition into a self-reinforcing activated immune state may occur. An entropy model can be utilized to consider the 'energy-barrier' to transition between immunosuppressive and activated immune states (Figure 4). The energy-barrier to transition between states may differ between highly immunogenic (left panel) and poorly immunogenic cancers (right panel). The molecular basis for differences between poorly immunogenic and highly immunogenic tumors are incompletely elucidated but new findings support a prominent contribution of mutational load in driving heterogeneous immunogenicity and clinical outcomes to immune-based therapy [107–109].

Figure 4



Bistable population dynamics of T_{reg} and T_{eff} cells within tumors. Entropy landscapes indicating bistable T_{reg} and T_{eff} cell population states in tumors. A stable low-entropy state involving functional predominance of T_{reg} cells over a minimal population of suppressed T_{eff} cells exists during the escape phase of tumor development (indicated by the position of the ball). Therapy (arrow) results in immune activation. *Left panel*: In the case of highly immunogenic tumors, an activation threshold is reached enabling stable transition into an activated immune state that results in tumor regression. *Right panel*: In the case of poorly immunogenic tumors, immune activation is insufficient to surmount the higher energy barrier to transition into an activated state resulting in stable persistence of immunosuppression and failure to clear disease.

A bistable model of T_{reg} and T_{eff} cell population dynamics has two important implications for cancer immunotherapy. First, the potential for immune-based therapies to achieve durable complete regression of disease distinguishes them from traditional chemotherapy. An attractive explanation for the striking durability of such clinical responses despite transient administration of therapy is the induction of self-reinforcing activation states within tumors. Secondly, while direct targeting of T_{reg} cells represents a strategy for disruption of immunosuppression in cancer, T_{eff} cell populations directly and indirectly antagonize T_{reg} cells. Thus, induction of immune activation represents an alternate strategy to reverse immunosuppression and a potential mechanism by which some therapies targeting T_{eff} cell function cause durable regression of disease. Thus, current immune-based therapies must be considered in the context of their effect on T_{reg} and T_{eff} cell population dynamics within tumors.

Attempts have been made to distinguish the effect of immune-based therapies on T_{reg} and T_{eff} cell populations within tumors. However, a majority of current immune-based therapies directly affect both T_{eff} and T_{reg} cells.

Monoclonal antibody therapy targeting CTLA-4 results in durable complete responses in patients with metastatic melanoma [6]. Initial reports attributed efficacy to an isolated effect on T_{eff} cell populations [110–112]. However, CTLA-4 treatment results in increased T_{eff}:T_{reg} cell ratios within tumors [113••,114,115] and its efficacy, in part, requires depletion of antibody-bound T_{reg} cells by Fcγ receptor–expressing macrophages within the tumor microenvironment [116,117]. Thus, CTLA-4 therapy both activates T_{eff} cells and depletes T_{reg} cells within tumors and it is plausible that disruption of the immunosuppressive balance of T_{reg} and T_{eff} cells contributes to durable tumor regression observed in a subset of treated patients. Similarly, monoclonal antibody therapy targeting the programmed cell death 1 (PD-1) receptor results in durable regression of disease in a substantial proportion of patients with metastatic melanoma [7••] and in a smaller proportion of patients with renal cell carcinoma, non-small cell lung cancer and ovarian cancer [8,118]. PD-1 is preferentially expressed on recently activated or exhausted T cells and negatively regulates effector function [119]. However, PD-1 signaling is also implicated in the induction and function of peripheral iT_{reg} cells [120] and blockade of PD-1 signaling causes both decreased $Foxp3^+$ T_{reg} cell ratios and augmented T_{eff} cell function in a murine melanoma therapy model [121]. Thus, the striking efficacy of monoclonal antibodies targeting PD-1 and CTLA-4 may be related to alterations in the balance of T_{reg} and T_{eff} cells within tumors. IL-12 also causes profound immune-mediated tumor regression with concomitant activation of T_{eff} cells in preclinical models, though systemic delivery of IL-12 in humans results in limiting adverse events [122,123]. In a number of mouse models, IL-12 administration causes increased T_{eff}:T_{reg} cell ratios [124,125]. While in some models, this is attributable to killing of T_{reg} cells by T_{eff} cells within tumors [90], IL-12 also has direct effects on the function and stability of T_{reg} cells [64].

In some cases, the effects of immune-based therapies on T_{reg}:T_{eff} ratios are unclear. High-dose IL-2 therapy results in durable clearance of metastatic melanoma and renal cell carcinoma in a minority of patients [126–128]. While this therapy results in transiently increased frequencies of peripherally circulating T_{reg} cells [129], its long-term effect on the ratio of T_{reg}:T_{eff} cells following withdrawal of therapy and specifically within tumors is not well established. In mice, administration of exogenous IL-2 drives differentiation of T_{eff} cells and loss of the IL-2-producing population of cells [50••]. This is significant given the function of IL-2-producing T_{eff} cells in maintenance of T_{reg} cell populations. Depletion of the IL-2 producing T_{eff} cell pool within tumors and loss of T_{reg} cell supportive capacity amongst TIL may represent a mechanism by which high-dose IL-2 causes durable complete regression of disease in a subset of patients. Durable complete responses are also observed following adoptive cell therapy (ACT) using bulk TIL populations [130–132] but the effect of therapy on endogenous T_{reg} and T_{eff} cell populations is poorly resolved. Reshaping of the endogenous T_{reg}:T_{eff} cell balance may, however, contribute to durable responses observed with both high-dose IL-2 therapy and ACT. Thus, a majority of immune-based therapies for cancer affect T_{eff} and T_{reg} cell population dynamics within tumors. Further detailed experimental investigation is required to separate direct from indirect effects of such therapies of T_{reg} and T_{eff} cell populations and their functional consequences.

In conclusion, mechanisms of reciprocal antagonism and self-reinforcement drive bistability in T_{reg} and T_{eff} cell population dynamics, enabling the immune system to exclusively consolidate the divergent outcomes of tolerance and immunity. An understanding of the complexity of T_{reg} and T_{eff} cell population dynamics has implications for rationalizing the striking durability and efficacy of immune-based therapies for cancer and provides a basis for development of new strategies that manipulate immune function in cancer patients.

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- • of special interest
- •• of outstanding interest

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