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The Potential Role of Regulatory T Lymphocytes (Tregs) In Cancer

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Review Article

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Abstract

Regulatory T cells (Tregs) are distinct type of T-cells which provide feedback control to any immune response. They are necessary to stop the immune response after the antigen has been successfully recognized. In the tumour micro-environment, the tumour often modulates the immune cells surrounding it in a way that it converts a large population of activated T-cells in to Tregs. For instance, tumours have been known to secrete IL-10 an inhibitory cytokine which is necessary for Treg formation. The tumour also modulates Dendritic cells (DC) and Macrophages so that they secrete inhibitory cytokines and promote tumorigenesis. Commonly, a Treg response to cancer cells, is to suppress the active immune response to the cancer. This review describes the recent studies of Treg cells in different human malignancies and discusses the restoration of antitumor immunity by depletion or reduced the functional strength of Treg cells hence, providing a promising tool to perfectly managing antitumor immune responses.

Keywords: Terg; IL-10; Tumour micro-environment; Activated T-cells

Abbreviations: DC: Dendritic Cells; Tregs: Regulatory T Cells; TME: Tumor Microenvironment; TAMs: Tumor-Associated Macrophages; HNSCC: Head and Neck Squamous Cell Carcinoma; CTLA4: Cytotoxic T Lymphocyte Antigen 4; CRC: Colorectal Carcinomas.

Introduction

The features of regulatory T (Treg) cells make them not only an added value and a significant milestone in the field of immunology but also provide an explanation for T-cell-mediated immunosuppression. T-cell-mediated immunosuppression [1-6]. An emerging evidence suggests that Treg cells play a significant role in tumor immunology and contribute to tumor growth and progression, thereby having a crucial prognostic value for cancer patients [7-12]. Peripheral blood immune cells include lymphocytes which are a complex type of leukocytes that direct the body's immune system. These cells are divided into three main types: (1) T helper cells: have CD4 (glycoproteins on the surface) which is recognized by MHC class II molecules on the antigen, (2) T cytotoxic cells: have CD8 which is recognized by MHC class I molecules on the antigen and (3) T regulatory cells: have CD4 and CD25 on the surface [13,14]. These are the negative regulators and act by suppressing the immune responses. Tregs appear to have a significant role in suppressing tumor-specific immunity. That is, they seem to prevent the immune system from attacking tumors [15]. Tregs, in general, prevent immune responses against "self-antigens" which are a part of one's own body. Accordingly, a Treg response to cancer cells, is to suppress the active immune response to the cancer. Autoimmunity is regarded as a potential outcome of tumor immunity, the mechanisms of autoimmunity and tumor immunity are associated [16].

Origin of Regulatory T Lymphocytes

Treg field was launched through the work of Sakaguchi, et al. [14] who in 1995 reported that a small group of T cells with particular cell surface phenotype (CD4+ cells which co-express the IL-2 receptor-α chain, CD25) maintain self-tolerance and that breakdown of this tolerance could lead to autoimmune disease and CD4+CD25+ T cells were named Treg cells. Now the terms "suppressor T cells" and "regulatory T cells" are used instead, but the term "regulatory T cells" is preferred by most researchers. Work in the field of Treg cell immunology was greatly enhanced in 2003 by the discovery and characterization of the Treg-specific gene, FoxP3 [17-23].

Regulatory T Lymphocytes and Helper T Lymphocytes

Regulatory T cells inhibit immune responses. Helper T cells tend to promote a given immune response by turning off other pathways. Interferons are classified as cytokines, small, secreted proteins that help regulate immune responses in a variety of ways. For example, a Th1 T helper cell will secrete IFN-gamma and will help to turn off Th2 responses and other non-Th1 responses. It will help provide IL-2 to cytotoxic T cells (CD8+ T cells) which is needed for survival. The T cell cytokine interleukin-2 (IL-2) is essential for the homeostasis of regulatory T (Treg) cells that suppress (auto) immunity and stimulates immune responses mediated by conventional T cells. Moreover, they will also license antigen presenting cells (APCs) to promote strong cytotoxic T cell responses and will allow the APCs to secrete polarization cytokines for a longer period of time, enhancing immune responses. The produced IFN-gamma can kill cancerous cells by means of perforins. These pore-forming proteins allow access for serine proteases called granzymes to the cytoplasm of the cell about to be killed. Granzymes also can initiate apoptosis through activation of caspases [24-28].

Treg and Tumor Microenvironment

Tumor cells are in tumor microenvironment (TME) able to shuffle off apoptosis and dodge immune surveillance [29,30]. The TME comprises of many immunosuppressive cells including regulatory T (Treg) cells, T helper type 2 (Th2) cells, tumor-associated macrophages (TAMs), myeloid-derived suppressor cells, and, in some case, Th17 cells [31-33]. Therefore, immunotherapy has a significant value to modulates host immune response and actively restores the specific anti-immunity to cancer. The tumour micro environment has a significant role in cancer development and metastases. The tumour is able to progressively change its surrounding environment and make it more prone to grow. There is a pretty recognition that tumour growth can be modulated by actively changing the tumour micro environment. Despite the Treg has the ability to maintain immune regulation it can be unfavorable in cancer through suppression of anti-tumor immunity [34-36]. Humans that lack a functional Treg population develop a lethal autoimmune disorder, termed Immune dysregulation. Certainly, high numbers of Tregs and a low CD8+ T cell: Treg ratio are considered poor prognostic factors for many tumor types, including melanoma, head and neck squamous cell carcinoma (HNSCC), ovarian cancer and colorectal carcinoma [37-41]. Accordingly, it has become apparent that the possibility of identifying the targeted pathways to reduce intratumoral Tregs, will provide a therapeutic approach in many patients with malignancy, while if a disruption occurs in the peripheral Treg number or function therefore, it will cause autoimmune diseases or inflammatory complications.

Foxp3+ Treg

Regulatory T (Treg) cells, with the master regulator Foxp3 (forkhead box P3), represent a functional distinct subset of CD4 T cells which make the Tregs able to do immune suppression and play a significant role in maintaining immune-homeostasis and autoimmune diseases [42-44]. Foxp3+ Treg cells exert their effector functions through a variety of molecular mechanisms. Firstly, Treg cells constitutively express the high-affinity heterotrimeric interleukin 2 (IL2) receptor, also known as CD25, which further bind to and consume IL2 from their surroundings, thus compromising its effects on non-Foxp3 effector T cells (Teff) [45,46]. Treg cells also express high level of cytotoxic T lymphocyte antigen 4 (CTLA4), which can bind to CD80/CD86 on antigen presenting cells (APCs) and thereby transmitting suppressive signals to these cells and reducing their capacity to activate Teff cells [47]. Besides, CTLA4 exhibits a higher affinity to CD80/CD86 than that of CD28, thus competing with this co-stimulatory receptor, which further disrupts the priming and/or activation of Teff cells [48]. Additionally, Treg cells can produce immunosuppressive cytokines, such as TGFβ, IL10, and IL35 [49], which will downregulate the activity of APCs and Teff cells, and secrete granzymes and perforin that can directly kill these cells [50]. Similarly, CD8 memory T cells consisting of effector memory T cells (TEM), central memory T cells (TCM) and tissue-resident memory T cells (TRM) [51]. Thymus derived Treg (tTreg) cells can also be divided as central (cTreg) and effector (eTreg) Treg cells based on the expression of trafficking receptors [52]. cTreg cells are programmed to recirculate through secondary lymphoid organs (SLOs) by expressing CD62L as well as CCR7, while eTreg cells capable of entering non-lymphoid tissues by virtue of expressing chemokine receptors such as CXCR3, CCR4, CCR6, CCR2, and CCR5, etc [53]. A number of adoptive transfer studies have reported that the TLR3L poly(I:C) is a potent adjuvant for CD8+ T cell responses, through

increasing T cell number, function, cytokine production, and anti-tumor response [54]. These adjuvant effects were associated with increases in the by increasing the numbers of dendritic cells in mice and in pancreatic cancer patients as well as increases in the numbers of NK cells in mice. In addition, in vivo administration of poly(I:C) at the peak of dendritic cell (DC) expansion after cyclophosphamide treatment induces inflammatory cytokine production and increases in the number of activated DCs in lymph nodes. Also, that poly(I:C) target CD8+ T cells directly and activate them in vitro, where adoptive transfer of these cells resulted in appreciated antigen-specific CD8+ T cell response and greater expansion in vivo. cTreg cells could convert into more highly proliferative eTreg cells in response to tissue self-antigens. Notably, eTreg cells have also been observed in increased numbers within diverse experimental mouse tumors and human cancers which suggest the involvement of Treg cells in anti-tumor immunity [55-58].

Treg cells are involved in tumor development and progression by inhibiting antitumor immunity. The Regulatory T (Treg) cells suppress abnormal/excessive immune responses to self- and nonself-antigens to maintain immune homeostasis. Regulatory T (Treg) cells expressing the transcription factor Foxp3 has an essential role in controlling autoimmunity and maintain immunological tolerance in mouse and human [59]. Treg cells can exert their suppressor function in non-lymphoid tissues as evidenced by specific tissue lesions in mice with selectively impaired Treg cell migration. In addition to secondary lymphoid organs, Treg cells can exert their suppressor function in non-lymphoid tissues as evidenced by specific tissue lesions in mice with selectively impaired Treg cell migration [60]. Treg cells have also been found in increased numbers in diverse experimental mouse tumors and in human cancers [61,62]. While breast carcinomas have not traditionally been considered immunogenic, evidence of tumor infiltrating lymphocytes and their subset composition paralleling disease progression suggest that the underlying interactions of these tumors with immune cells are important. An increased ratio of CD4+ to CD8+ T cells correlates with lymph node metastases and reduced overall survival [63]. Increased presence of Treg cells in breast tumor biopsies is associated with an invasive phenotype and diminished relapse-free as well as overall survival this suggesting a reduced antitumor specific immunity by the action of the Treg cells [64-66].

Prognostic Value of Tumor-Infiltrating T Regulatory Cells (TI-Tregs)

Numerous studies reported the presence of unlimited number of Treg cells in various malignancies including, hepatocellular, gastric, lung, breast, ovarian, cervical, and melanomas [67-73] which reflecting the unfavorable prognosis in these tumors. On the contrary, other studies reported a favorable role of FOXP3+ T-cells in colorectal carcinomas (CRC) [74-76]. The investigators explained the significance of CRC favorable outcomes is primarily due to the presence of higher expression of FOXP3 which is indeed infiltrated more with FOXP3loCD45RA+effector T-cells and upregulated inflammatory genes such as Il12a, Il12b, Tgfb1, and Tnf while, in other tumors a higher infiltration of FOXP3hiCD45RA− cells resulted in poor prognosis and lower disease-free survival [77].

Treg and Impact of Interleukin 10 (IL-10)

Monocytes which are usually found in the blood can differentiate into either a macrophage or a dendritic cell. Based on chemical signaling from the nearby tissues, the monocyte can change into the required cell type. Dendritic cells (DCs) have the ability to control the immune response through induction and polarization of primary, antigen-specific immune responses. According to their maturation/activation status, their surface molecules, and the produced cytokines the DCs are either to evoke immune responses by activation of effector T cells or to induce tolerance by the induction of either T cell anergy, regulatory T cells, or production of regulatory cytokines. Among the cytokines produced by tolerogenic DCs, from one hand interleukin 10 (IL-10) is a key regulatory cytokine limiting excessive T-cell responses to pathogens to prevent chronic inflammation and tissue damage also interleukin-10 (IL-10) acts as a poor prognostic marker in many cancers [78,79].

IL-10 is primarily produced by monocytes and to a lesser extent by type 2 T helper cells (TH2), mastocytes, CD4+CD25+Foxp3+ regulatory T cells, and certain subsets of activated T cells and B cells [80]. IL-10 can inhibit the synthesis of pro-inflammatory cytokines such as IFN-γ, IL-2, IL-3 and TNF-α produced by cells such as Mφ and regulatory T-cells4. Moreover, IL-10 can act on regulatory T cells to maintain transcription factor Foxp3 expression and suppressive functions in mice with colitis [81]. Regulatory T cells (Tregs) are present throughout the body and have. Tregs are able to prevent autoimmunity and immunopathology and maintaining immunological homeostasis suggesting their crucial role in immunity [82,83]. The role of IL-10 in cancer is to facilitate tumor immune escape suggesting that IL-10 in the tumor-microenvironment of different cancers is correlated with poor prognosis [84].

Depletion of Treg Cells and Restoration of Antitumor Immunity in Human Tumor Progression

The specific immunity against tumor antigen could be improved through depletion of circulating

CD4+CD25highFoxP3+ Treg cells after multiple doses of immunotoxin, denileukin diftitox thus suggesting an encouraging outcome for combined Treg depletion and anticancer vaccines [85]. The experiments demonstrated that, the Treg cells in spleen, peripheral blood, and bone marrow of normal C57BL/6 mice were variously reduced after a single intraperitoneal injection of denileukin diftitox [86]. The reduction was peaked within 24 hours and continued almost for 10 days following injection. Injection of denileukin diftitox 1 day before vaccination has a positive impact on anticancer immunity evidenced by antigen-specific T cell responses above levels induced by vaccination alone. Therefore, the Treg cell depletion improves endogenous anti-tumor immunity and the efficacy of active immunotherapy in animal models for cancer, suggesting that inhibiting Treg cell function could also improve the limited successes of human cancer immunotherapy [87]. In one study, authors reported that removal of Treg cells in tumor-bearing mice improved the function of adoptively transferred cells for new antitumor immunotherapies [88]. In other studies, authors allowing Treg cells to migrate to sites of disease more efficiently than effector cells include CCR4, CCR5, CCR6, CCR7 and CCR8 [89,90]. They demonstrated that CCR5-dependent chemotaxis is essential for Treg cell migration into pancreatic adenocarinoma but if a disruption takes place to the signaling it would result in a decrease in migration of Treg cell into the tumor, allowing a decline in tumor growth [90]. Thereby, Treg cell migration with its collateral signaling into tumor microenvironment is considered a novel immunomodulatory strategy for the treatment of cancer.

Conclusion

The occurrence of Treg cells marked as (CD4+CD25+FoxP3+ T cells) have recently gained prominence through their prevalence in numerous malignancies. The broad growing understanding of the immune regulation is closely associated with the recognition of FOX-P3 which represents the receptor of Treg cells thus, tumor tissue helps to turn the naive T cells into FoxP3 Treg cells and accumulates Treg cells in tumor site, therefore declining the development of anti-tumor specific immunity. Nevertheless, the depletion of CD4+CD25+ Treg cells contributes to the restoration of the antitumor immunity through promoting the activation of immune T cells in the draining lymph nodes, thus, facilitating adoptive immunotherapy. The tumour often modulates the immune cells surrounding it in a way that it converts a large population of activated T-cells in to Treg cells. For instance, tumours have been known to secrete IL-10 an inhibitory cytokine which is necessary for Treg formation. The significance behind depletion of Treg cells is to abundantly reveal the T- cell antitumor activity and ligation of CD3 and CD28 enhances both innate (NKT cells) and adaptive (CD4 + and CD8+ T cells) responses to develop an antitumor micro-environment which is able to suppress tumor growth.

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