

## The role of T regulatory cells in asthma

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As a chronic inflammatory disease, much of the research related to asthma has been focused on proinflammatory mechanisms. Recently, advances have been made in defining mechanisms that control inflammation and induce immune tolerance to specific antigens. Subsets of CD4<sup>+</sup> cells known as T regulatory cells play an important role in directing these processes, and recent experiments have begun to define crucial molecular and signaling pathways. There is a growing body of evidence describing the function of T regulatory cells in the development, disease activity, and treatment responses related to asthma and other atopic diseases. Collectively, this new information suggests that a greater understanding of these pathways might lead to new therapeutic targets for asthma and other diseases of chronic airway inflammation. (*J Allergy Clin Immunol* 2005;116:996-9.)

**Key words:** Asthma, T regulatory cells, inflammation, CD25, Foxp3, tolerance

### ASTHMA AND REGULATION OF AIRWAY INFLAMMATION

The recognition of asthma as a chronic inflammatory disease in the 1980s led to new paradigms for asthma pathogenesis and treatment. Experiments in mice identified specific CD4<sup>+</sup> T cells, known as T<sub>H</sub>2 cells, which produce cytokines that regulate IgE production and eosinophilic inflammation; these cells were later found to be overrepresented in the airways of asthmatic individuals. This led to the concept that T<sub>H</sub>2 skewing of proinflammatory immune responses plays a fundamental role in the development of asthma and other atopic diseases. Although this concept is still valid, therapies directed at inhibiting T<sub>H</sub>2 processes in mouse models and in clinical

#### Abbreviations used

APC: Antigen-presenting cell

CTLA4: Cytotoxic T-lymphocyte antigen 4

Treg: T regulatory

trials have generally met with only partial success. Furthermore, there is substantial evidence that the inflammatory processes in the asthmatic airway leading to airway dysfunction are not limited to T<sub>H</sub>2 cells.<sup>1</sup> For example, viral infections, which are classic stimulators of T<sub>H</sub>1 immune responses, cause asthma-like symptoms in infancy, might contribute to the initiation of asthma, and commonly cause exacerbations of this disease. In addition, diseases related to asthma, such as chronic atopic dermatitis, have components of both T<sub>H</sub>1 and T<sub>H</sub>2 inflammation. These observations suggest that an impaired ability to regulate inflammation in general could be an important contributing factor in asthma and perhaps other atopic disorders. The search for immunoregulatory mechanisms has recently centered on a specific subset of T regulatory cells (CD25<sup>+</sup> Treg cells) that serve to limit inflammatory responses and to promote immune tolerance in a variety of experimental systems and clinical disorders.

### OVERVIEW OF CD25<sup>+</sup> Treg CELLS

Historically, the evidence for a population of T cells that actively suppress immune responses in a dominant fashion dates back to the 1960s. The path to discovery of this elusive T-cell subset has had several setbacks and is still an area of investigation filled with unresolved issues. Nonetheless, discoveries made by Sakaguchi et al<sup>2</sup> have clarified and firmly established the existence of a CD25-bearing CD4<sup>+</sup> T-cell subset that serves an essential role in the maintenance of homeostasis and peripheral T-cell tolerance to self-antigens. These “naturally occurring” Treg cells represent a distinct lineage of T cells that develop in the thymus and bear a broad repertoire of self-reactive T-cell receptors.<sup>3</sup> A second variety of Treg cells can be induced in the peripheral lymphoid tissues in response to foreign antigens or allergens (Fig 1).<sup>4,5</sup> The

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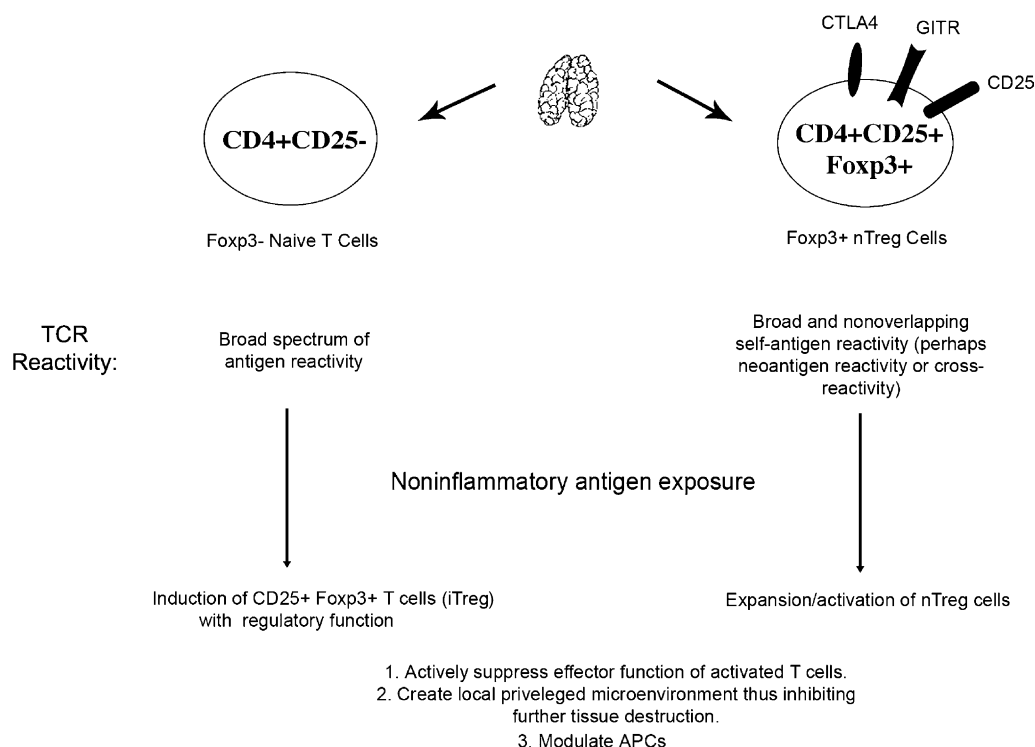


**FIG 1.** Derivation of naturally occurring and induced CD25<sup>+</sup> Treg cells after exposure to cognate antigen in a noninflammatory environment. *GITR*, Glucocorticoid-induced TNF receptor–related gene; *TCR*, T-cell antigen receptor; *nTreg*, naturally occurring (or thymically derived) CD25<sup>+</sup> Treg cell; *iTreg*, induced (or adaptive) CD25<sup>+</sup> Treg cell.

adaptive CD25<sup>+</sup> Treg cells develop when antigen is presented in noninflammatory conditions. A better understanding of the development and mechanisms of adaptive CD25<sup>+</sup> Treg cells will have important implications for allergic diseases and asthma.

One of the problems in studying Treg cells is the lack of a unique set of cell-surface markers. Receptors associated with Treg cells, such as CD25, cytotoxic T-lymphocyte antigen 4 (CTLA4), and glucocorticoid-induced TNF receptor family–related receptor, are also expressed on antigen-activated CD4<sup>+</sup> T cells. To date, the only unique genetic marker for CD25<sup>+</sup> Treg cells is the transcription repressor factor Foxp3. Foxp3 is believed to be a master regulator of CD25<sup>+</sup> Treg cell development and function. In fact, Foxp3 mutations in mice and human subjects cause immune dysregulation and organ-specific autoimmune disease. Although its function remains elusive, Foxp3 is believed to repress the expression of various proinflammatory cytokine genes, and enforced expression in CD4<sup>+</sup> T cells is sufficient for conversion to a suppressor phenotype.<sup>6</sup>

### CD25<sup>+</sup> Treg CELLS: MECHANISM OF ACTION

The mechanism of action of CD25<sup>+</sup> Treg cells remains an area of intense investigation and ongoing controversy.

CD25<sup>+</sup> Treg cells can suppress a broad range of cell-mediated immune processes, suggesting a regulatory role in multiple disease states, including allergy.<sup>7</sup> A variety of model systems have implicated several potential mechanisms of action; however, to date, a unifying or widely accepted pathway for suppressor activity has not been defined, and it seems probable that there might be several nonredundant mechanisms (Fig 2). Most experimental evidence indicates that CD25<sup>+</sup> Treg cells require cell contact, engagement of their T-cell receptor, and IL-2 for their suppressor biologic function to be conveyed. CD25<sup>+</sup> Treg cells can directly suppress CD25<sup>−</sup> T cells or indirectly cause suppression through effects on antigen-presenting cells (APCs).<sup>7</sup>

TGF- $\beta$  is a complex cytokine that has been arguably implicated to be central in the biologic function of CD25<sup>+</sup> Treg cells.<sup>8</sup> Membrane-associated TGF- $\beta$  on the surface of CD25<sup>+</sup> Treg cells could be important in suppressor activity mediated by cell contact.<sup>8</sup> Recent provocative studies have demonstrated that exposure to high doses of TGF- $\beta$  upregulates Foxp3 in naive CD4<sup>+</sup> T cells.<sup>9,10</sup> Furthermore, there is evidence that TGF- $\beta$  signaling is necessary for cell suppression,<sup>11,12</sup> although this is not without controversy.<sup>13</sup>

CTLA4 is a surface receptor that is increased on CD25<sup>+</sup> Treg cells and might be important for suppressor function. CTLA4 could act as an inhibitory molecule on engagement of CD80 or CD86 on effector T cells or APCs.<sup>7</sup>

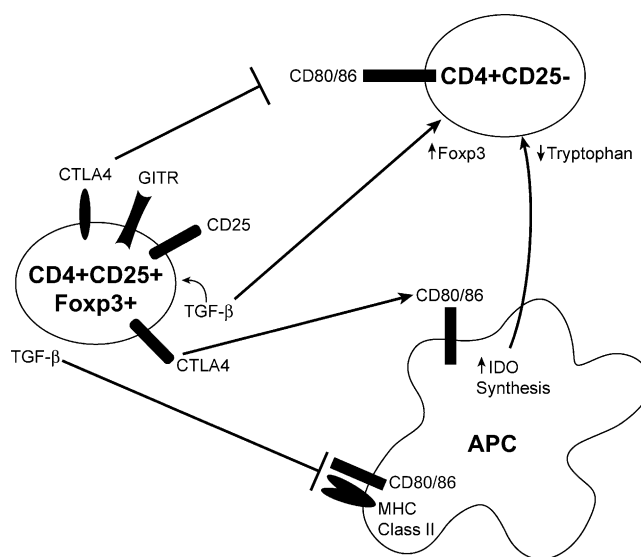


**FIG 2.** Potential mechanisms for CD25<sup>+</sup> Treg cell suppression of T-cell and APC functions. *IDO*, Indoleamine 2,3-dioxygenase.

This binding can induce APCs to produce indoleamine 2,3-dioxygenase and catabolize tryptophan, resulting in diminished T-cell proliferation.<sup>14</sup>

## EVIDENCE FOR A ROLE OF CD25<sup>+</sup> Treg CELLS IN ALLERGIC DISEASE

Mutations in the gene encoding Foxp3 are associated with an absence of CD25<sup>+</sup> Treg cells and are the underlying cause of the X-linked immune dysregulation, polyendocrinopathy, enteropathy, and X-linked (IPEX) syndrome, which is characterized by polyendocrinopathy, enteropathy, and immune dysregulation. Notably, patients with IPEX syndrome typically have eczema, eosinophilia, increased serum IgE levels, and enhanced T<sub>H</sub>2 responses.<sup>15</sup> Thus there is conclusive evidence that Foxp3 and CD25<sup>+</sup> Treg cells play a role in suppression of T<sub>H</sub>2-driven immune responses in human subjects, and similar findings have been described in mice.

Several recent reports have begun to investigate the role of CD25<sup>+</sup> Treg cells in common allergic diseases. Although these studies do not provide a direct causal relationship between allergic inflammation and CD25<sup>+</sup> Treg cells, they do provide a strong argument for ongoing exploration. CD25<sup>+</sup> Treg suppressor function was studied in seasonal atopic patients compared with nonatopic control subjects in and out of their pollen season by Ling et al.<sup>16</sup> Regardless of the exposure to environmental allergens, CD25<sup>+</sup> Treg cells from the atopic group were less able to suppress polyclonal T-cell proliferation compared with those from the nonatopic control subjects. A direct causal relationship between Foxp3 expression and suppression of allergic immune responses remains to be demonstrated. However, a recent descriptive report demonstrated that glucocorticoids upregulate

Foxp3 expression.<sup>17</sup> This suggests that one operational function of glucocorticoids might be to increase the gene expression of Foxp3 in T cells and skew the immune response toward immunosuppression, thus restoring balance to dysregulated allergic inflammatory responses.

Using a murine model of asthma, Doganci et al<sup>18</sup> recently provided evidence that the diminished presence of CD25<sup>+</sup> Treg cells resulted in increased airway hyper-reactivity, thus providing strong evidence linking CD25<sup>+</sup> Treg cells to airway physiology. In addition, the results of this study indicate that the proinflammatory cytokine IL-6 might play a direct role in the local presence of CD25<sup>+</sup> Treg cells. For example, blocking IL-6 in the airway resulted in increased numbers of lung CD25<sup>+</sup> Treg cells that expressed Foxp3 and exhibited immunosuppressive capabilities. The source of IL-6 was not explored in this study; however, it is tempting to speculate that lung APCs are responsible for directing the development, expansion, or both of CD25<sup>+</sup> Treg cells. In support of this hypothesis, engagement of Toll-like receptors on APCs can induce production of IL-6 and abrogate CD25<sup>+</sup> Treg suppressor activity.<sup>19</sup> When considered together, these findings link APC production of IL-6 to the suppression of CD25<sup>+</sup> Treg development in the lung and potentially provide an avenue for therapeutic intervention in asthma.

In summary, it has become clear over the past few years that the accepted paradigm of allergic disease and asthma being caused by T<sub>H</sub>2-skewed adaptive immune responses is an oversimplification. Recent advances in the field of immunology have broadened our understanding of how adaptive immune responses are regulated: there is mounting evidence that CD25<sup>+</sup> Treg cells are an important component of this process. Although there are presently more questions than answers, the investiga-

tion of CD25<sup>+</sup> Treg cells in regulating inflammatory responses and immune tolerance is fertile for important and therapy-altering discoveries related to allergic diseases.

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