Pinning Allergies on pathogenic Th2 cells

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Abstract

A new pro-inflammatory subtype of antigen-specific Th2 cell that expresses CD161 emerges as the pathogenic cell type in allergic disease and is deleted during allergen-specific immunotherapy (Wambre et al.) (1).

Allergic diseases including asthma, allergic rhinitis (hay fever), atopic dermatitis (eczema), food allergy and anaphylaxis are major health concerns with increasing prevalence particularly in the “developed” world. These diseases affect a considerable proportion of the population, result in considerable morbidity, and are in some circumstances life-threatening. Asthma affects around 300 million people worldwide and kills several thousand people each year in the United States alone, and anaphylaxis is frequently life threatening for sufferers although the use of adrenaline/epinephrine auto-injectors has improved this situation in some countries. It has long been known that being atopic (allergic) and having one allergic disease increases the likelihood of developing other allergic diseases during a person’s life time - the so-called atopic march. Allergic diseases are driven by an inappropriate immune response to normally innocuous antigens such as cat dander, peanuts, grass pollen or house dust mite. Despite intensive research we still do not fully understand why some individuals develop antigen-specific pathogenic immune responses to these allergens. Almost as mysterious are the T cells that drive the allergic response. In this issue, Wambre et al. (1) bring us a step closer to identifying and understanding this enigmatic T cell. They show that a new subset of pathogenic Th2 cells (Th2A) is found specifically in patients with allergic disease and link depletion of Th2A cells with allergen desensitisation immunotherapy.
Th2 cells in allergic disease

A number of elegant studies in both mouse and human have shown that the pathology of allergic disease is driven by the type 2 cytokines, which induce different hallmarks of allergic disease: IL-4 induces B-cell isotype switching to produce IgE, IL-5 promotes eosinophil differentiation, migration and survival, and IL-13 induces mucus hypersecretion and fibrosis. Type 2 cytokines can be produced by a variety of innate immune cell types including mast cells, basophils and the more recently identified group 2 innate lymphoid cells (ILC2s). However, antigen-specific expression of Type 2 cytokines in response to allergen is restricted to the Th2 subset of T-helper cells.

The Th2 cytokines are not constitutively expressed by Th2 cells but are transiently activated and secreted upon antigen stimulation, making identification of the Th2 cell subset in the absence of specific allergen challenging. In humans Th2 cells can be identified by expression of the prostaglandin D₂ receptor CRTh2 (also known as DP2 or CD294). Paradoxically however, numerous studies have failed to detect an increased prevalence of CRTh2⁺ Th2 cells in patients with allergic disease. In fact, recent studies have suggested that there is functional heterogeneity within the Th2 cell subset with some memory Th2 cell populations being able to express high levels of IL-5 (2, 3), IL-17 (4) or IFNγ (5) alongside IL-4 and IL-13, giving rise to the concept of a pathogenic effector Th2 (peTh2 or Tpath2) cell subset (6). The nature of this Th2 cell subset and the methods used to identify it are an area of active research with several potential phenotypic markers being proposed in both mouse and human studies. These markers include the receptors for the epithelial derived cytokines IL-25 (IL17RB) (7), IL-33 (ST2) (8) and Thymic Stromal Lymphopoietin (TSLP receptor; TSLPR)(9). These epithelial-derived cytokines are believed to be released from damaged epithelium in response to viral infection, allergen exposure or physical damage, and potently induce Type 2 cytokine production from a variety of cell types including ILC2s, basophils, mast cells and Th2 cells.

Identification of pathogenic Th2 cells

In their new study, Wambre et al. (1) identified allergen-specific T-cells in patients by selecting patients with allergy to alder tree pollen, using peptides from alder allergens in combination with fluorescently labelled major histocompatibility class II (MHC II) tetramers.
This is how an allergen-specific T-cell would normally ‘see’ an allergen on the surface of an antigen presenting cell and therefore allowed the investigators to truly identify the T-cells responsible for the response to a particular allergen. They used this approach to screen a large panel of cell surface molecules in an attempt to identify markers that are selectively expressed on allergen specific T-helper cells. As expected, they found that most allergen-specific T-cells were CRTh2+ and had a memory phenotype (CCR7lowCD27lowCD45RBlow). Crucially however, they also identified two surface markers that were selectively upregulated in the allergen-specific Th2 cell population, CD161 and CD49d. The expression of the C-type lectin receptor CD161 is closely associated with inflammatory Th17 cells, however the authors convincingly demonstrate that these CD27-CD45RB- CRTh2+CD161+CD49d+ Th2 cells do not express hallmarks of Th17 differentiation including IL-17 and RORγt, distinguishing these cells from those observed by Cosmi et al.(4). They went on to show that in patients with a wide range of allergies including food (peanut), perennial (cat), seasonal pollen (Timothy grass) and mold (Aspergillus), antigen-specific Th2 cells with the CD27-CD45RB-CRTh2+CD161+CD49d+ phenotype were present at high levels, whilst they were absent in non-allergic people, suggesting that these cells may be causing allergic disease.

Although identifying allergen-specific Th2 cells that may cause disease is a big step forward, the authors’ approach requires prior knowledge of the allergen involved and the ability to develop specific peptide MHCII tetramers against the allergen. From a clinical viewpoint, this may not always be possible and would thus restrict the usefulness of this approach. However, Wambre and colleagues went on to see whether they could identify this Th2 cell population in patients without using the MHCII tetramer technology. They found that compared with non-allergic people, allergic individuals had higher frequencies of these pathogenic Th2 cells, dubbed “Th2A cells” and defined as CD27 CD45RB- CRTh2+CD161+CD49d+ (Fig 1).

To attempt to demonstrate that the pathogenic Th2 cells were indeed the cells that respond to allergen in atopic individuals they examined patients undergoing two different allergen challenge situations: natural exposure of hay fever sufferers during the pollen season and a double-blind placebo controlled peanut challenge in patients with peanut allergy. In both situations, the pathogenic Th2 cell subset (CD27 CRTh2+CD161+) was activated in response
to in vivo allergen challenge, as assessed by expression of the activation marker CD38, in contrast to the conventional Th2 cell population (CRTh2+CD161+). Thus, the authors appear to have identified the allergen responsive pathogenic Th2 cell population in humans and report that it is a subset of the total Th2 cell pool.

Allergen desensitisation immunotherapy is a mainstay of treatment for several allergic diseases including allergic rhinitis and bee venom anaphylaxis although the precise mechanisms of action are not fully understood. Patients are administered either subcutaneously or sublingually increasing amounts of allergen to desensitise them. Wambre et al. went on to examine the impact of a peanut immunotherapy treatment on their pathogenic Th2 cell subset. Remarkably, they found that the pathogenic Th2 cell population was selectively deleted in patients undergoing the active immunotherapy treatment and that this specifically correlated with clinical benefit, suggesting that pathogenic Th2 cells are a clinically relevant target cell population in allergic disease.

**Mechanistic insights**

To explore the mechanisms by which pathogenic Th2 cells might cause disease they performed multi-parametric intracellular cytokine staining and demonstrated that pathogenic Th2 cells expressed more IL-5 and IL-9 than conventional Th2 cells and do not express IL-17 or IFN-γ. Transcriptome profiling of the pathogenic Th2 cells identified an interesting set of genes upregulated in this population including the IL-25 receptor (IL17RB), the IL-33 receptor (ST2; IL1RL1) and TSLPR (CLRF2), Cox-2 (PTGS2) and hematopoietic prostaglandin D synthase (HPGDS). Collectively this gene expression profile appears very similar to the various pathogenic effector Th2 cell populations identified by several other groups. We have previously identified IL17RB+ST2+ Th2 cells in nasal polyposis (7) and others have found HPGDS+peTh2 cells in patients with eosinophilic gastrointestinal disease and atopic dermatitis (9). Wambre et al. failed to detect surface expression of IL17RB or ST2 in their experiments possibly due to the use of different detection antibodies. It is highly likely that all of these studies have been examining the same or very similar pathogenic effector Th2 cell subsets which provides significant hope that therapies targeting this cell population will provide benefit in a wide variety of allergic diseases.
One outstanding question regarding pathogenic Th2 cells surrounds the mechanism of differentiation compared to conventional Th2 cells. Classical Th2 and pathogenic Th2 cells express GATA3, the master regulator of Th2 differentiation. It will be important to determine whether additional transcription factors are involved in directing the pathogenic Th2 transcriptional program; one candidate for this could be the nuclear receptor PPARγ reported to be upregulated in pathogenic Th2 cells by Wambre et al. It will also be important to understand the role of the epithelial derived cytokines in regulating pathogenic Th2 cell function and activation.

**Clinical Implications**

The identification of pathogenic Th2 cells as a clinically important cell type in allergic disease will enable researchers to use this reported panel of cell surface markers to study the pathogenic Th2 response to novel treatments for these various diseases. The Wambre et al. study is timely since several pharmaceutical companies have new therapeutics targeting IL-25, IL-33, TSLP and their respective receptors (IL17RB, ST2 and TSLPR) entering clinical programs. Furthermore, given that pathogenic Th2 cells express both the biosynthetic pathway for PGD₂ production (Cox-2 and HPGDS) and a receptor for PGD₂ (CRTh2), it will be critical to examine the effect of blockade of this pathway in allergic disease. The recent study of the drug Fevipiprant in asthma, a small molecule antagonist of CRTh2, suggests that CRTh2 antagonism may be one mechanism worth exploring in other allergic diseases(10).

Overall the study by Wambre et al. implicates a new pathogenic Th2 cell subset in allergic disease, and reveals a panel of cell surface markers that the research community can utilise to examine the importance of these pathogenic Th2 cells in clinical trials of new therapeutic agents.

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**References**


Figure Legend

**Figure 1. Pathogenic effector Th2 cells in the spotlight.** Allergen challenge to the epithelium allows dendritic cells to present allergen to pathogenic Th2 cells and induces release of epithelial-derived cytokines such as IL-25, IL-33 and TSLP. Pathogenic Th2 cells are allergen-specific CD4⁺ T-cells identified by a panel of cell surface markers (CD27 CD45RB⁻ CRTh2⁺CD161⁺CD49d⁺) and epithelial cytokine receptors (IL17RB, ST2 and TSLPR). Pathogenic Th2 cells are activated by allergen and produce type 2 cytokines and PGD₂ in response to epithelial cytokines. CRTh2 antagonists and antibodies targeting epithelial cytokines may block pathogenic Th2 cell recruitment or activation. These pathogenic Th2 cells that drive allergen-specific immune responses may be a useful therapeutic target for treating allergic disease.
Epithelium

Dendritic cell

MHCII

TCR

CRTh2 antagonist

Pathogenic effector

Th2/Th2A cell

Activated pathogenic effector Th2/Th2A cell

Anti IL-25

Anti IL-33

Anti TSLP

IL-25

IL-33

TSLP

IL-4

IL-5

IL-9

IL-13

PGD2

IL-17RB

ST2

TSLPR

CRTh2

GATA3

PPARG

PTGS2

CRTh2 antagonist

PGD2

CD161

Pathogenic effector Th2/Th2A cell

Activated pathogenic effector Th2/Th2A cell

Allergen