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## Innate type 2 cells and asthma

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Asthma and other allergic conditions are immunologically described as Th2 skewed, with associated increases in IgE, eosinophils and expression of IL-4, IL-5 and IL-13. However, recent advances have implicated other type 2 cytokines such as IL-25, IL-33 and TSLP in the pathogenesis of allergic conditions. These cytokines are potent inducers of the panel of recently described innate immune cells, which also produce large amounts of the Th2 cytokines IL-5 and IL-13 independent of the adaptive immune response. We review herein, the role for these innate cell populations with a focus on the nuocyte, in allergic asthma and pulmonary inflammation.

### Addresses

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### Introduction

Asthma is a serious chronic inflammatory lung disease affecting approximately 300 million people worldwide associated with increased mortality in patients, which can be characterized as a spectrum of pulmonary conditions underpinned by airway inflammation that leads to airway hyper-responsiveness (AHR). Clinical asthma is a polygenic disease with recent genome-wide association studies (GWAS) implicating a number of gene associations [1], the complexity of which suggests that studies to elucidate the interplay between environmental factors and implicated genes will require extensive investigation. The heterogeneity of asthma is such that multiple immune pathways are implicated in both the aetiology and pathogenesis of the disease. A minimalist division of the disease spectrum is into intrinsic asthma (associated with aberrant response to allergen and the development of atopy) and extrinsic (not allergen driven or atopic but associated with a range of environmental factors). Intrinsic allergic asthma has been the attention of the majority of research to-date. Allergic asthma shares similarities

with most atopic conditions, such as allergic rhinitis, atopic dermatitis and anaphylaxis, in that it generally occurs in childhood and 'episodes' of disease exacerbation are attributed to an aberrant hyperactive CD4<sup>+</sup> helper 2 (Th2) T cell cytokine response to common innocuous allergens. The generation of multiple allergic conditions within an individual can be attributed to the atopic march, whereby clinical signs of allergic conditions occur sequentially persisting over a number of years, typically originally manifesting in early childhood with allergic eczema preceding the development of allergic asthma and rhinitis. The stereotypical allergic immune response is initiated by activation of allergen-specific Th2 cells, release of the Th2 cytokines Interleukin (IL)-4, IL-5, IL-9 and IL-13 coupled with an increase in levels of IgE and localized eosinophilia [2]. The resulting pathological response in allergic asthma includes lung inflammation, oedema, increased mucus production and smooth muscle contraction, resulting in episodes of allergen induced AHR, eventually resulting in lung damage and fibrosis [3]. Although a predominant amount of research has been conducted into Th2 cell-mediated 'allergic' asthma, consistent with a heterogeneous condition other non T cell responses have a growing recognition as contributing to genesis of allergic-like inflammation [4]. While traditional cells such as macrophages, basophils and eosinophils are implicated in disease pathogenesis, the emergence of new innate cells opens new avenues for asthma research.

The incidence of allergic conditions is now rising worldwide, with the prevalence of asthma in particular increasing by 1% every year [5]. Many hypotheses have been suggested for the rise in incidence of allergic conditions in recent years. Among these is the long-standing 'hygiene hypothesis', which suggests that improvements in public health care, the use of vaccines and antibiotics and a consequential alteration in mucosal microbiota have resulted in decreased childhood exposure to pathogens, such as bacteria, viruses and parasites [6,7]. Exposure of the innate immune system to such pathogens in early life may beneficially alter the regulatory response to infection, which, if unchallenged may give rise to development of either Th2-mediated allergic responses or Th1/Th17-mediated autoimmune conditions [8]. In the context of the debate on the rise of asthma in modern society, recent studies have implied that, contrary to prior belief based on traditional bacterial culture techniques, the respiratory tract is not a sterile environment and that the pulmonary microbial flora may differ between health and disease. Thus using polymorphic bacterial *16S-rRNA* gene screening of lungs the presence of certain bacterial strains in the

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lungs of children and adults with asthma was identified [9]. The lung is therefore subject to the elegant interplay between the full gamut of innate immune recognition including Toll-like receptors (TLRs) interactions with pathogen-associated molecular patterns (PAMPs) and also endogenous danger-associated molecular patterns (DAMPs). Indeed inherent to asthma initiation are the interface between stimuli and lung dendritic cells [10] and the interplay with the epithelium [11]. The known lower incidence of asthma in children living on farms, relative to urban dwellers, has now been shown to be associated with exposure to a complex of microorganisms in early childhood in such children [12], highlighting the link between gene-environment in asthma.

Of particular interest in understanding the incidence of allergic conditions, is the concurrent decrease in endemic infection with parasitic helminths, in particular in developing countries. In murine studies, the parasitic helminths such as *Schistosoma mansoni*, *Nippostrongylus brasiliensis* and *Trichuris muris* have proved invaluable in dissecting the Th2 response, both with regard to the progression of parasitic infection, but also concerning the interplay between infection and allergy [13]. Recent studies have dissected the mechanisms underpinning innate activation of a Th2 response during such helminth infections and identified subsets of unique type 2 innate cells, which will be discussed herein. Current studies have now progressed from the formal identification of cell phenotypes to reporting a role for such cells in disease conditions affecting mucosal tissues, with particular reference to inflammatory conditions in the lung [14<sup>••</sup>,15–18]. Running concurrently with murine studies, several recent publications also report the presence of novel innate type 1 and 2 cells in humans, with Mjosberg *et al.* and Monticelli *et al.*, reporting a population of innate type 2 cells present in the chronic type 2 condition, rhinosinusitis [19] and as playing a role in influenza [17]. Outlined in this review are the recent developments in understanding the innate activation of T cell responses and how these newly identified cells may affect the progression of allergic conditions, with particular reference to the role of the nuocyte in the pathogenesis of allergic asthma.

### Type 2 immunity in allergy and asthma

Allergic asthma occurs as a result of exposure to environmental airborne allergens, such as house dust mites and animal dander, many of which are unable to act directly on CD4<sup>+</sup> T cells and therefore induce Th2 cell differentiation through intermediate mediators [20]. Activation of a Th2 response is typically characterized by localized and systemic activation of the cytokines IL-4, IL-5, IL-9 and IL-13, which, in turn induce antibody class switching to IgG1 and IgE. Th2 cytokines recruits type 2 effector cells such as eosinophils, mast cells and basophils, resulting in goblet cell hyperplasia and mucus production as well as alternative-activation of macrophages. The

importance in asthma of the archetypal Th2 cytokines in particular IL-4, IL-5 and IL-13 are well documented. IL-4 promotes the production of IgE by B cells, IL-5 activates eosinophil development and recruitment to the lung, while IL-13 is thought to mediate airway remodeling and hyperresponsiveness. IL-5 was one of the first Th2 cytokines to be investigated as a target for treating asthma with anti-IL-5 antibodies altering eosinophil numbers in patient tissue but exerting limited overall clinical impact. This has lead to strategies to target the IL-5 receptor [21]. More recently, developments in the clinical treatment of allergic asthma are refocused on blocking IL-4 and IL-13 signaling [3]. Targeting IL-4 has thus far proved unsuccessful with the use of soluble recombinant IL-4R $\alpha$  (altrakinecept) and a humanized anti-IL-4 monoclonal antibody (mAb; pascolizumab) failing in patient clinical trials [22,23]. However, the use of anti-IL-13 mAbs is showing promise in early stage allergen response studies and several studies are still in progress using anti-IL-13 mAbs (QAX576, CAT-354, MILR1444A) in combination with traditional corticosteroid treatment regimes [2,24]. Collectively, targeting the classic Th2-associated cytokines has shown limited success, partly owing to the heterogeneous nature of asthma but also owing to our incomplete understanding of the disease process. Therefore other new therapeutic targets for asthma are required.

In addition to the traditional Th2 cytokines IL-4, IL-5, IL-9 and IL-13, non-lymphoid derived cytokines such as TSLP (thymic stromal lymphopoietin), IL-25 (IL-17E) and IL-33 are also widely recognized as integral in promoting Th2 differentiation. The role for TSLP in asthma was highlighted in mouse models, with TSLP over expression leading to increased AHR, while TSLP-deficient mice show impaired development of type 2 responses in ovalbumin-induced airway inflammation models [25]. In addition a recent report from Shikotra *et al.*, demonstrates increased expression of immunoreactive TSLP in both the airway epithelium and bronchial lamina propria of patients with severe asthma [26]. IL-33 is a member of the IL-1 family and signals through T1/ST2 (encoded by *Il1r1*) [27]. Administration of IL-33 to mice induces typical type 2 responses, including localized eosinophilia, IgE class switching and increased mucus production, while blocking IL-33-induced signaling results in decreased AHR [27,28]. Similar to IL-33, administration of IL-25 induces type 2 responses, furthermore, blocking IL-25 either by using IL-25 deficient mice or an IL-25 neutralising antibody decreases the severity of AHR in ovalbumin-induced airway inflammation models [29–31]. IL-25 utilises the receptor IL-17RB in both mice and humans. It has been identified that polymorphisms in *IL-17RB* in humans can be linked with asthma [32]. Of importance is the ability of both IL-25 and IL-33 to induce expression of IL-5 and IL-13 from non-B/non-T cells (NBNT) [33,34<sup>•</sup>,35<sup>•</sup>], see

below. Studies using T and B lymphocyte deficient *Rag2*<sup>-/-</sup> mice showed that during helminth infection, both IL-25 and IL-33 are able to induce IL-5 and IL-13, suggesting a NBNT source of these cytokines [29,34<sup>•</sup>,35<sup>•</sup>]. In addition to type 2 cytokines, IL-17 and IL-22 have also been implicated in the pathogenesis of asthma [36]. Patients with severe asthma demonstrate increased presence of neutrophils in the broncho-alveolar lavage (BAL) fluid than patients with mild disease [37]. IL-17A is closely associated with the increase in neutrophils and interestingly, in addition to increased presence of Th17 cells in the airways of asthmatic patients, IL-17A may directly alter airway smooth muscles cells and thereby influence AHR [38,39].

### Innate cells in type 2 immunity

The mechanisms regulating activation of the innate immune system with regard to viral and bacterial stimulation are well known, by contrast the mechanisms governing the innate activation of Th2-mediated responses to allergy and helminth infections are only recently being elucidated. For example, in the last 2 years four studies identified novel NBNT cells that are initiated in mice in response to helminth infections. These novel NBNT cells have been shown to play a role in the generation and pathology of allergic conditions [34<sup>•</sup>,35<sup>•</sup>,40<sup>•</sup>,41<sup>•</sup>]. In 2006 a NBNT cell population elicited by IL-25 and/or IL-33 and devoid of any lineage markers was described [33]. The cells did not express FcεR1, CD49b or CCR3, and thus are not mature basophils, mast cells or eosinophils (Table 1). Absence of these lineage negative populations render the mice unable to expel the parasite *N. brasiliensis*

so is therefore important for a competent type 2 immune response. Further analysis of the cells identified by Fallon *et al.*, resulted in identification of the nuocyte reported by Neill *et al.* in 2010. The nuocyte was identified based on its expression of IL-13 and can be further defined as lineage<sup>-</sup>ICOS<sup>+</sup>CD45<sup>+</sup>T1/ST2<sup>+</sup>IL-17BR<sup>+</sup>. In addition to upregulation during *N. brasiliensis* infection, nuocytes can also be induced by intra-peritoneal injection of IL-25 or IL-33 [34<sup>•</sup>].

Moro *et al.* identified a similar innate cell population they termed the natural helper (NH) cell [35<sup>•</sup>]. The NH cells are identified as lineage<sup>-</sup> but express c-Kit, Sca-1, CD25, CD127 and T1/ST2 and are found largely in fat-associated lymphoid clusters (FALC), which are located along the gut mesentery of mice surrounded by adipose tissue [35<sup>•</sup>]. In addition to IL-13, NH cells produce IL-5 and IL-6 constitutively, which is upregulated by stimulation with IL-33 or a combination of IL-25 and IL-2 [35<sup>•</sup>]. What was particularly original about NH cells was the demonstration that they were also present in human FALC [35<sup>•</sup>]. Using *T. muris* the Artis group identified an innate murine type 2 cell population responsive to IL-25, termed multipotent progenitor (MPP)<sup>type2</sup> cell [40<sup>•</sup>]. Although studies using *T. muris* have identified this cell as important in the type 2 immune response and it shares phenotypic similarities with the other type 2 innate cells, this cell type is unique in its ability to differentiate in culture to mast cells or basophils [40<sup>•</sup>]. In addition, Locksley and colleagues described innate helper 2 (Ih2) cells as an additional IL-25 and IL-33-responsive population [41<sup>•</sup>]. These cells are widely distributed in the mouse, with

**Table 1**

**A comparison between mouse and human innate cells. (MLN: mesenteric lymph node, FALC: fat associated lymphoid clusters, BM: bone marrow, GALT: gut associated lymphoid tissue)**

|                                                           | Mouse                                                           |                                                                                     |                                                           |                                                                                                  | Human                                   |                                         |
|-----------------------------------------------------------|-----------------------------------------------------------------|-------------------------------------------------------------------------------------|-----------------------------------------------------------|--------------------------------------------------------------------------------------------------|-----------------------------------------|-----------------------------------------|
|                                                           | Nuocytes                                                        | NH cells                                                                            | Ih2 cells                                                 | MPP <sup>type2</sup>                                                                             | CD127 <sup>+</sup> ILC                  | CRTH2 <sup>+</sup> type 2 ILC           |
| Anatomical location                                       | MLN, spleen, peritoneum, lung                                   | FALC                                                                                | MLN, spleen, liver, BM                                    | MLN, GALT                                                                                        | Gut                                     | Gut, lung, peripheral blood             |
| Surface phenotype                                         |                                                                 |                                                                                     |                                                           |                                                                                                  |                                         |                                         |
| Lineage <sup>-</sup> c-kit <sup>+</sup> Sca1 <sup>+</sup> | Yes                                                             | Yes                                                                                 | Lineage <sup>-</sup> c-kit <sup>+</sup> Sca1 <sup>-</sup> | Yes                                                                                              | Lineage <sup>-</sup> c-kit <sup>+</sup> | Lineage <sup>-</sup> c-kit <sup>+</sup> |
| T1/ST2 <sup>+</sup>                                       | Yes                                                             | Yes                                                                                 | Unknown                                                   | No                                                                                               | Unknown                                 | <i>IL1r1</i> mRNA expression            |
| IL-7Ra <sup>+</sup>                                       | Yes                                                             | Yes                                                                                 | Unknown                                                   | No                                                                                               | Yes                                     | Yes                                     |
| CD44 <sup>+</sup>                                         | Yes                                                             | Yes                                                                                 | Yes                                                       | No                                                                                               | Unknown                                 | Unknown                                 |
| CD90.1 <sup>+</sup>                                       | Yes                                                             | Yes                                                                                 | Yes                                                       | No                                                                                               | Unknown                                 | Unknown                                 |
| Transcription factor expression                           | <i>RORA</i>                                                     | <i>Maf</i> , <i>Gata-3</i> , <i>JunB</i> , <i>Stat6</i> , <i>Id2</i> , <i>Tbx21</i> | <i>Gata-3</i> , <i>Stat6</i>                              |                                                                                                  | <i>ID2</i> , <i>RORC</i>                | <i>Id2</i>                              |
| Cytokine responsivity                                     | IL-25, IL-33                                                    | IL-25, IL-33                                                                        | IL-25, IL-33                                              | IL-25                                                                                            | IL-2, IL-7, IL-15                       | IL-25, IL-33                            |
| Physiological significance                                | Secrete IL-5 and IL-13<br>Increase goblet cell mucus production | Secrete IL-5 and IL-13<br>Increase goblet cell mucus production                     | Secrete IL-5 and IL-13<br>Increase eosinophilia           | Upregulate <i>Il4</i> , <i>Il5</i> and <i>Il13</i> mRNA<br>Increase goblet cell mucus production | Secrete IL-22, IL-17                    | Secrete IL-5 and IL-13                  |

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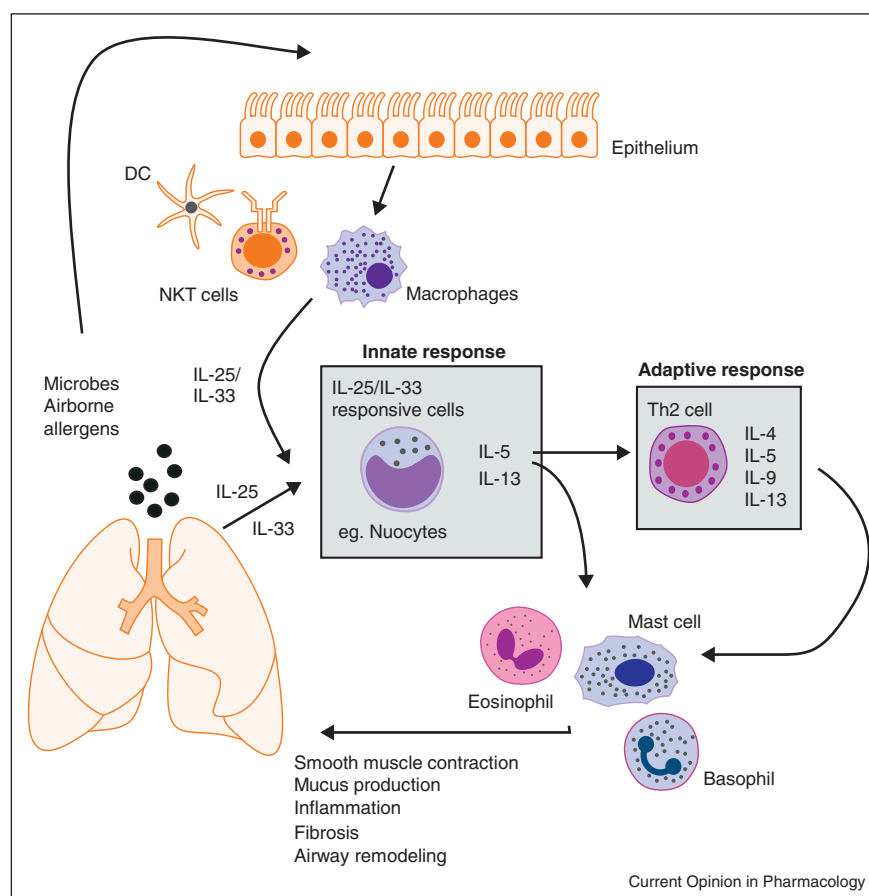
expression in the mesenteric lymph node, spleen, liver and bone marrow upregulated on days 5–7 post *N. brasiliensis* infection [41<sup>•</sup>].

Although all these 4 cell types described are phenotypically and morphologically similar, analysis of the surface markers indicates these cells are defined (Table 1). For example, while the nuocyte and NH cell express both T1/ST2 and CD127, MPP<sup>type2</sup> cells do not. In addition, unlike the nuocyte, the NH cell does not express ICOS, but expresses CD25. Further to their distinct surface marker expression, each cell population differs in expression of transcription factors. The MPP<sup>type2</sup> cells appear to be less differentiated than Ih2 cells, NH cells and nuocytes, not expressing mRNA encoding GATA-3, c-Maf, STAT6 or JunB [40<sup>•</sup>]. By contrast, Ih2 cells express *Gata3* and *Stat6* [41<sup>•</sup>], while NH cells express *c-Maf*, *Gata3* and *Stat6* [35<sup>•</sup>]. More recent work has shown that nuocytes develop from common lymphoid progenitors (CLPs) via IL-33, IL-7 and Notch dependent pathways [42<sup>••</sup>]. Retinoid orphan

nuclear receptor alpha (ROR $\alpha$ ) has been identified as the dominant transcription factor in the nuocyte [42<sup>••</sup>], which is of particular interest as genetic polymorphisms in the *RORA* gene have been associated with asthma in humans [1].

Further to these initial studies in mice, the identification of innate cells has been noted in humans (Table 1). These cells have been grouped under the title of lymphoid tissue inducer cells (LTi) or innate lymphoid cells (ILC), and the population originally identified express CD117, CD127 and the transcription factor RORC, while not expressing lineage markers, and produce IL-17 and IL-22 [43]. These and other populations subsequently identified share many characteristics with NK cells and are present in human tonsils, intestinal lamina propria, colon and Peyer's patches and play an important role in protection against bacterial infection [44]. A type 2 innate lymphoid cell has recently been identified in humans which if lineage<sup>−</sup>CD127<sup>+</sup> and can be characterized by

**Figure 1**



Innate activation of a type 2 response in the lung. Inhaled airborne allergens can trigger a type 2 response in the lung by activation of innate type 2 cells such as the nuocyte. Allergens trigger the release of IL-25 and IL-33 from airway epithelial cells activating the innate type 2 cells to release the Th2 cytokines IL-5 and IL-13. In addition to activating Th2 cells, these cytokines also activate mast cells and eosinophils that leads to airway smooth muscle contraction, goblet cell hyperplasia and mucus production and airway inflammation.

expression of the Th2 marker chemoattractant receptor-homologous molecule expressed on Th2 lymphocytes (CRTH2) [19]. These cells were found in human mucosal tissues as well as peripheral blood and, similar to the murine innate type 2 cells, produce IL-5 and IL-13 on stimulation with IL-25 and IL-33 [19].

### The nuocyte, lung associated innate cells and airway inflammation

While nuocytes were originally defined using an *Il13* reporter mouse as lineage<sup>−</sup>CD45<sup>+</sup>T1/ST2<sup>+</sup>IL-17BR<sup>+</sup>, they also express ICOS, Sca1, CD90.2 (Thy1.2), CD117 (c-kit),  $\beta_7$  and CCR9 [34<sup>•</sup>]. As nuocytes have been shown to be potent producers of IL-13, and IL-13 is essential for development of AHR, mucus production and inflammation, recent studies have focused on the potential for nuocytes to play a role in allergic lung inflammation [14<sup>••</sup>]. Further to identification of nuocyte in gut associated mucosal tissue during *N. brasiliensis* infection, work by Barlow *et al.* has shown expression of the nuocyte in both the BAL and lung tissue in response to ovalbumin-induced allergic lung inflammation [14<sup>••</sup>]. Using IL-4 and IL-13 reporter mice, nuocytes were identified as a potent innate source of IL-13, with transfer of nuocytes able to restore AHR and eosinophilia in cytokine-induced lung inflammation in *Il13* deficient mice [14<sup>••</sup>].

In addition to the nuocyte, another IL-33-responsive innate lymphoid cell has been identified as resident in the lung of naïve mice. These cells share similar receptor expression to the nuocyte, they are lineage<sup>−</sup> but express CD25, CD44, CD90.2, CD127, Sca1 and ICOS, however, unlike the nuocyte, these cells do not express c-kit or IL-17RB, so are unresponsive to IL-25 [15]. Upon stimulation with exogenous IL-33 or in response to the common airborne allergen *Alternaria alternata*, these cells produce IL-5 and IL-13 and are necessary for lung eosinophilia and AHR [15]. A unique population of innate IL-5 producing cells have also been reported in the lung of naïve mice, which share several phenotypic similarities with the NH cell [16]. Although these cells

were identified using a tumour metastasis model, they also demonstrate the ability to regulate eosinophil infiltration into the lung [16]. Concurrently with these studies were additional publications reporting populations of ILCs, similar to nuocytes and NH cells in the airway mucosal tissue of mice [18,45<sup>••</sup>] and an analogous population found in human BAL [17]. The mouse lung ILCs were shown to play a crucial role in regulating airway epithelial integrity and orchestrating pulmonary tissue homeostasis after experimental infection with influenza virus [17] and are critical for induction of eosinophilic lung inflammation in response to cysteine proteases [45<sup>••</sup>].

These recent studies outline the presence of innate type 2 cells in the lung and demonstrate through their expression and production of the Th2 cytokines IL-5 and IL-13 a role in allergic inflammation in the lung (Figure 1). All cell types investigated are able to affect lung eosinophilia, which is vital in the pathogenesis of clinical allergic asthma. In addition, both the nuocyte and the IL-33-responsive innate lymphoid cell were essential in the pathogenesis of AHR. The presence of these innate cells in the lung, and their observed importance in regulating airway type 2 responses and homeostasis, is of significance when evaluating the potential of the innate cells in the initiation of asthma.

### Conclusions

The identification of a multitude of type 2 innate cell populations both in mice and in humans outlined herein provide further insight on the innate mechanisms underlying type 2 immunity. These cells, either present in the mucosal tissues of naïve mice, or stimulated by release of the Th2 cytokines IL-25 and IL-33, clearly play an important role in the generation of a type 2 immune response, as outlined in studies using helminth infection. Of real interest is the presence of several of these cell populations, notably the nuocyte, in the lung during allergic lung inflammation models, suggesting the potential for these cells to play a role in asthma or other

**Table 2**

#### Expression of genes associated with asthma in mouse and human type 2 cells

| Loci with genomewide significance for asthma | Presence in type 2 cells |                               |             |           |
|----------------------------------------------|--------------------------|-------------------------------|-------------|-----------|
|                                              | Innate                   |                               | Adaptive    |           |
|                                              | Mouse                    | Human                         | Mouse/human |           |
| Gene                                         | Nuocytes                 | CRTH2 <sup>+</sup> type 2 ILC | Eosinophils | Th2 cells |
| <i>IL18R1</i>                                | –                        | –                             | Yes         | No        |
| <i>HLA-DQ</i>                                | –                        | –                             | No          | No        |
| <i>IL33</i>                                  | –                        | –                             | No          | No        |
| <i>SMAD3</i>                                 | –                        | –                             | Yes         | Yes       |
| <i>IL2RB</i>                                 | –                        | Yes                           | –           | Yes       |
| <i>IL13</i>                                  | Yes                      | Yes                           | Yes         | Yes       |
| <i>RORA</i>                                  | Yes                      | –                             | –           | No        |

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pulmonary disorders. Supporting these observations was a recent genome-wide association study, which identified polymorphisms in the genes for IL-13, IL-33 and the transcription factor RORA as significant in a cohort of patients with asthma [46\*\*] (Table 2). These data taken in combination outline the importance of recent discoveries in innate activation of type 2 responses. New innate type 2 cells and their associated cytokines have transformed current paradigms on the initiation and development of asthma and have novel potential to be therapeutically targeted in asthma or other allergic conditions.

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## References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- of special interest
- of outstanding interest

1. Zhang Y, Moffatt MF, Cookson WO: **Genetic and genomic approaches to asthma: new insights for the origins.** *Curr Opin Pulm Med* 2012, **18**:6-13.
2. Gauvreau GM, Boulet LP, Cockcroft DW, Fitzgerald JM, Carlsten C, Davis BE, Deschesnes F, Duong M, Durn BL, Howie KJ *et al.*: **Effects of interleukin-13 blockade on allergen-induced airway responses in mild atopic asthma.** *Am J Respir Crit Care Med* 2011, **183**:1007-1014.
3. Holgate ST: **Pathophysiology of asthma: what has our current understanding taught us about new therapeutic approaches?** *J Allergy Clin Immunol* 2011, **128**:495-505.
4. Kim HY, Chang YJ, Subramanian S, Lee HH, Albacker LA, Matangkasombut P, Savage PB, McKenzie AN, Smith DE, Rottman JB *et al.*: **Innate lymphoid cells responding to IL-33 mediate airway hyperreactivity independently of adaptive immunity.** *J Allergy Clin Immunol* 2011, **129**:216-227 e211-216.
5. Braman SS: **The global burden of asthma.** *Chest* 2006, **130**:4S-12S.
6. Strachan DP: **Hay fever, hygiene, and household size.** *BMJ* 1989, **299**:1259-1260.
7. Wills-Karp M, Santeliz J, Karp CL: **The germless theory of allergic disease: revisiting the hygiene hypothesis.** *Nat Rev Immunol* 2001, **1**:69-75.
8. Ngoi SM, Sylvester FA, Vella AT: **The role of microbial byproducts in protection against immunological disorders and the hygiene hypothesis.** *Discov Med* 2011, **12**:405-412.
9. Hilty M, Burke C, Pedro H, Cardenas P, Bush A, Bossley C, Davies J, Ervine A, Poulter L, Pachter L *et al.*: **Disordered microbial communities in asthmatic airways.** *PLoS ONE* 2010, **5**:e8578.
10. Lambrecht BN, Hammad H: **Lung dendritic cells in respiratory viral infection and asthma: from protection to immunopathology.** *Annu Rev Immunol* 2012, **30**:243-270.
11. Lloyd CM, Saglani S: **Asthma and allergy: the emerging epithelium.** *Nat Med* 2010, **16**:273-274.
12. Ege MJ, Mayer M, Normand AC, Genuneit J, Cookson WO, Braun-Fahrlander C, Heederik D, Piarroux R, von Mutius E: **Exposure to environmental microorganisms and childhood asthma.** *N Engl J Med* 2011, **364**:701-709.
13. Fallon PG, Mangan NE: **Suppression of TH2-type allergic reactions by helminth infection.** *Nat Rev Immunol* 2007, **7**:220-230.
14. Barlow JL, Bellosi A, Hardman CS, Drynan LF, Wong SH, Cruickshank JP, McKenzie AN: **Innate IL-13-producing nuocytes arise during allergic lung inflammation and contribute to airways hyperreactivity.** *J Allergy Clin Immunol* 2011, **129**:191-198 e194.
15. Bartemes KR, Iijima K, Kobayashi T, Kephart GM, McKenzie AN, Kita H: **IL-33-responsive lineage-CD25+ CD44hi lymphoid cells mediate innate type 2 immunity and allergic inflammation in the lungs.** *J Immunol* 2011, **188**:1503-1513.
16. Ikutani M, Yanagibashi T, Ogasawara M, Tsuneyama K, Yamamoto S, Hattori Y, Kouro T, Itakura A, Nagai Y, Takaki S *et al.*: **Identification of innate IL-5-producing cells and their role in lung eosinophil regulation and antitumor immunity.** *J Immunol* 2011, **188**:703-713.
17. Monticelli LA, Sonnenberg GF, Abt MC, Alenghat T, Ziegler CG, Doering TA, Angelosanto JM, Laidlaw BJ, Yang CY, Sathaliyawala T *et al.*: **Innate lymphoid cells promote lung-tissue homeostasis after infection with influenza virus.** *Nat Immunol* 2011, **12**:1045-1054.
18. Chang YJ, Kim HY, Albacker LA, Baumgarth N, McKenzie AN, Smith DE, Dekruyff RH, Umetsu DT: **Innate lymphoid cells mediate influenza-induced airway hyper-reactivity independently of adaptive immunity.** *Nat Immunol* 2011, **12**:631-638.
19. Mjosberg JM, Trifari S, Crellin NK, Peters CP, van Drunen CM, Piet B, Fokkens WJ, Cupedo T, Spits H: **Human IL-25- and IL-33-responsive type 2 innate lymphoid cells are defined by expression of CCR2 and CD161.** *Nat Immunol* 2011, **12**:1055-1062.
20. Oliphant CJ, Barlow JL, McKenzie AN: **Insights into the initiation of type 2 immune responses.** *Immunology* 2011, **134**:378-385.
21. Molino NA, Gossage D, Kolbeck R, Parker JM, Geba GP: **Molecular and clinical rationale for therapeutic targeting of interleukin-5 and its receptor.** *Clin Exp Allergy* 2012, **42**:712-737.
22. Borish LC, Nelson HS, Corren J, Bensch G, Busse WW, Whitmore JB, Agosti JM: **Efficacy of soluble IL-4 receptor for the treatment of adults with asthma.** *J Allergy Clin Immunol* 2001, **107**:963-970.
23. Hart TK, Blackburn MN, Brigham-Burke M, Dede K, Al-Mahdi N, Zia-Amirhosseini P, Cook RM: **Preclinical efficacy and safety of pascalizumab (SB 240683): a humanized anti-interleukin-4 antibody with therapeutic potential in asthma.** *Clin Exp Immunol* 2002, **130**:93-100.
24. Oh CK, Faggioni R, Jin F, Roskos LK, Wang B, Birrell C, Wilson R, Molino NA: **An open-label, single-dose bioavailability study of the pharmacokinetics of CAT-354 after subcutaneous and intravenous administration in healthy males.** *Br J Clin Pharmacol* 2010, **69**:645-655.
25. Zhou B, Comeau MR, De Smedt T, Liggitt HD, Dahl ME, Lewis DB, Gyarmati D, Aye T, Campbell DJ, Ziegler SF: **Thymic stromal lymphopoietin as a key initiator of allergic airway inflammation in mice.** *Nat Immunol* 2005, **6**:1047-1053.
26. Shikotra A, Choy DF, Ohri CM, Doran E, Butler C, Hargadon B, Shelley M, Abbas AR, Austin CD, Jackman J *et al.*: **Increased expression of immunoreactive thymic stromal lymphopoietin in patients with severe asthma.** *J Allergy Clin Immunol* 2011, **129**:104-111 e109.
27. Schmitz J, Owyang A, Oldham E, Song Y, Murphy E, McClanahan TK, Zurawski G, Moshrefi M, Qin J, Li X *et al.*: **IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines.** *Immunity* 2005, **23**:479-490.
28. Oboki K, Ohno T, Kajiwara N, Arae K, Morita H, Ishii A, Nambu A, Abe T, Kiyonari H, Matsumoto K *et al.*: **IL-33 is a crucial amplifier of innate rather than acquired immunity.** *Proc Natl Acad Sci USA* 2010, **107**:18581-18586.

29. Fort MM, Cheung J, Yen D, Li J, Zurawski SM, Lo S, Menon S, Clifford T, Hunte B, Lesley R *et al.*: **IL-25 induces IL-4, IL-5, and IL-13 and Th2-associated pathologies in vivo.** *Immunity* 2001, **15**:985-995.
30. Angkasekwinai P, Park H, Wang YH, Chang SH, Corry DB, Liu YJ, Zhu Z, Dong C: **Interleukin 25 promotes the initiation of proallergic type 2 responses.** *J Exp Med* 2007, **204**:1509-1517.
31. Ballantyne SJ, Barlow JL, Jolin HE, Nath P, Williams AS, Chung KF, Sturton G, Wong SH, McKenzie AN: **Blocking IL-25 prevents airway hyperresponsiveness in allergic asthma.** *J Allergy Clin Immunol* 2007, **120**:1324-1331.
32. Jung JS, Park BL, Cheong HS, Bae JS, Kim JH, Chang HS, Rhim T, Park JS, Jang AS, Lee YM *et al.*: **Association of IL-17RB gene polymorphism with asthma.** *Chest* 2009, **135**:1173-1180.
33. Fallon PG, Ballantyne SJ, Mangan NE, Barlow JL, Dasvarma A, Hewett DR, McIlgorm A, Jolin HE, McKenzie AN: **Identification of an interleukin (IL)-25-dependent cell population that provides IL-4, IL-5, and IL-13 at the onset of helminth expulsion.** *J Exp Med* 2006, **203**:1105-1116.
34. Neill DR, Wong SH, Bellosi A, Flynn RJ, Daly M, Langford TK,
  - Bucks K, Kane CM, Fallon PG, Pannell R *et al.*: **Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity.** *Nature* 2010, **464**:1367-1370.

This article is the first to identify the nuocyte using IL-13eGFP reporter mice and outlines the importance of IL-25 in the expansion of the nuocyte, demonstrating a severe defect in the ability of IL-25 deficient mice to expel parasite that can be rescued by the transfer of IL-13 producing nuocytes.
35. Moro K, Yamada T, Tanabe M, Takeuchi T, Ikawa T, Kawamoto H,
  - Furusawa J, Ohtani M, Fujii H, Koyasu S: **Innate production of T(H)2 cytokines by adipose tissue-associated c-Kit(+)Sca-1(+) lymphoid cells.** *Nature* 2010, **463**:540-544.

This article identifies innate lymphoid cells present in a novel lymphoid structure associated with adipose tissue in the peritoneal cavity.
36. Souwer Y, Szegedi K, Kapsenberg ML, de Jong EC: **IL-17 and IL-22 in atopic allergic disease.** *Curr Opin Immunol* 2010, **22**:821-826.
37. Wenzel SE, Szefer SJ, Leung DY, Sloan SI, Rex MD, Martin RJ: **Bronchoscopic evaluation of severe asthma. Persistent inflammation associated with high dose glucocorticoids.** *Am J Respir Crit Care Med* 1997, **156**:737-743.
38. Wang YH, Voo KS, Liu B, Chen CY, Uygungil B, Spoede W, Bernstein JA, Huston DP, Liu YJ: **A novel subset of CD4(+) T(H)2 memory/effector cells that produce inflammatory IL-17 cytokine and promote the exacerbation of chronic allergic asthma.** *J Exp Med* 2010, **207**:2479-2491.
39. Kudo M, Melton AC, Chen C, Engler MB, Huang KE, Ren X, Wang Y, Bernstein X, Li JT, Atabai K *et al.*: **IL-17A produced by alphabeta T cells drives airway hyper-responsiveness in mice and enhances mouse and human airway smooth muscle contraction.** *Nat Med* 2012, **18**:547-554.
40. Saenz SA, Siracusa MC, Perrigoue JG, Spencer SP, Urban JF Jr,
  - Tocker JE, Budelsky AL, Kleinschek MA, Kastelein RA, Kambayashi T *et al.*: **IL25 elicits a multipotent progenitor cell population that promotes T(H)2 cytokine responses.** *Nature* 2010, **464**:1362-1366.

This article describes how IL-25 can induce a population of lineage negative progenitor cells in gut associated lymphoid tissue. These multipotent progenitor cells can give rise to cells of monocyte/macrophage or granulocyte lineage both *in vitro* or *in vivo*.
41. Price AE, Liang HE, Sullivan BM, Reinhardt RL, Eisle CJ, Erle DJ,
  - Locksley RM: **Systemically dispersed innate IL-13-expressing cells in type 2 immunity.** *Proc Natl Acad Sci USA* 2010, **107**:11489-11494.

This study uses IL-4 and IL-13 reporter mice to define an innate population of cells that arise in response to IL-25 and IL-33 *in vivo*. These cells are widely dispersed and a major producers of IL-5 and IL-13 during infection with *N. brasiliensis*.
42. Wong SH, Walker JA, Jolin HE, Drynan LF, Hams E, Camelo A,
  - Barlow JL, Neill DR, Panova V, Koch U *et al.*: **Transcription factor RORalpha is critical for nuocyte development.** *Nat Immunol* 2010, **13**:229-236.

This study demonstrates the importance of the transcription factor RORalpha in the development of the nuocyte and their role in the expulsion of parasitic worms.
43. Cupedo T, Crellin NK, Papazian N, Rombouts EJ, Weijer K, Grogan JL, Fibbe WE, Cornelissen JJ, Spits H: **Human fetal lymphoid tissue-inducer cells are interleukin 17-producing precursors to RORC+ CD127+ natural killer-like cells.** *Nat Immunol* 2009, **10**:66-74.
44. Cella M, Fuchs A, Vermi W, Facchetti F, Otero K, Lennerz JK, Doherty JM, Mills JC, Colonna M: **A human natural killer cell subset provides an innate source of IL-22 for mucosal immunity.** *Nature* 2009, **457**:722-725.
45. Halim TY, Krauss RH, Sun AC, Takei F: **Lung natural helper cells are a critical source of Th2 cell-type cytokines in protease allergen-induced airway inflammation.** *Immunity* 2012, **36**:451-463.

This recent article describes the presence of innate lymphocytes, termed lung natural helper cells, as a source of Th2 cytokines in the lungs of mice treated with the cysteine protease papain.

46. Moffatt MF, Gut IG, Demenais F, Strachan DP, Bouzigon E,
 

- Heath S, von Mutius E, Farrall M, Lathrop M, Cookson WO: **A large-scale, consortium-based genomewide association study of asthma.** *N Engl J Med* 2011, **363**:1211-1221.

This GWAS study shows genomewide significance between asthma and single-nucleotide polymorphisms involving IL1R1, IL33 and RORA.