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INVITED REVIEW

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Innate and adaptive type 2 immunity in lung allergic inflammation

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Summary

Allergic inflammation is a type 2 immune disorder classically characterized by high levels of immunoglobulin E (IgE) and the development of Th2 cells. Asthma is a pulmonary allergic inflammatory disease resulting in bronchial hyper-reactivity. Atopic asthma is defined by IgE antibody-mediated mast cell degranulation, while in non-atopic asthma there is no allergen-specific IgE and more involvement of innate immune cells, such as basophils, group 2 innate lymphoid cells (ILC2), and eosinophils. Recently, protease allergens were shown to cause asthmatic responses in the absence of Th2 cells, suggesting that an innate cell network (IL-33/TSLP-basophil-ILC2-IL-5/IL-13 axis) can facilitate the sensitization phase of type 2 inflammatory responses. Recent evidence also indicates that in the chronic phase, these innate immune cells directly or indirectly contribute to the adaptive Th2 cell responses. In this review, we discuss the role of Th2 cytokines (IL-4 and IL-13) and innate immune cells (mast cells, basophils, ILC2s, and dendritic cells) in the cross-talk between innate and adaptive inflammatory responses.

KEYWORDS
Allergy, Asthma, Cell Lineages and Subsets, Immune-mediated Diseases, Mast Cells/Basophils, Th1/Th2/Th17 Cells, Tissues

1 | INTRODUCTION

Type 2 immune responses to allergens and parasitic infection are characterized by the presence of Th2 cells and immunoglobulin E (IgE). The type 2 response contributes to allergic inflammatory disorders, including allergic asthma, allergic rhinitis, atopic dermatitis (AD), and anaphylaxis. Repeated exposure to persistent allergen leads to chronic allergic inflammation. Asthma is a long-term pulmonary allergic inflammatory disease resulting in airway hyperresponsiveness (AHR), mucus hypersecretion, airway remodeling, and irreversible airflow obstruction. The asthmatic reaction can be provoked by specific allergen as well as by non-specific stimuli like cold air in exercise-induced asthma. The disease is very common, worldwide approximately 300 million people are afflicted. The number of people with asthma continues to grow. In the United States, 8.6% of children and 7.4% of adults had asthma in 2014, and in Japan, 11-14% of children and 6-10% of adults had asthma in 2015.

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Asthma was originally defined as a type 2-mediated allergic inflammatory disease that promotes barrier defenses at mucosal surfaces. The type 2 responses are regulated by various cytokines, such as IL-4, IL-5, IL-9, and IL-13.[1,2] Th2 cell-derived IL-4 is thought to promote IgE production by B cells in response to a particular allergen, and the allergen-specific IgE antibodies bind to the high-affinity Fcε receptor 1 (FcεR1) on basophils and mast cells. The allergen-specific IgE antibodies can be detected by measuring their levels in serum by ELISA and derivative techniques, or by a skin prick test in which a small amount of allergen is placed on or below the skin to see if a reaction develops. IgE antibodies to substances including house dust mite (HDM), skin dander from dog and cat, basidiomycete cap, mycelia, or spores of fungi, plant or tree pollen, and foods (e.g. wheat, peanuts, eggs, milk, fish, and shellfish) have been detected by these methods. IgE crosslinking on basophils and mast cells activates and releases a variety of mediators to control inflammation through degranulation. On the other hand, IL-5 and IL-9 are independently responsible for eosinophilia and mast cell hyperplasia. IL-13 regulates goblet cell proliferation, followed by the induction of mucin and mucus production, a hallmark of allergic asthma.[3–5]

Conventional asthma mouse models have clearly indicated the importance of Th2 cells, IgE antibody-mediated mast cell degranulation, and eosinophilic inflammation (Figure 1).[1] In contrast, non-atopic asthma is defined by the absence of serum IgE antibody reactive to allergens and more involvement of innate immune cells, such as group 2 innate lymphoid cells (ILC2), basophils, and eosinophils (Figure 2). Non-allergic asthma inflammation is induced by allergen proteases, including those from papain and HDM. The HDM-derived protease Derp1 causes the airway influx of eosinophils and bronchoconstriction in asthma patients.[6] The plant-derived cysteine protease, papain, causes occupational asthma.[7] The protease activity of papain causes epithelial cell barrier disruption and the stress of tissue injury leads to the elevation of damage-associated cytokines, including IL-25, IL-33, and thymic stromal lymphopoietin (TSLP). These proteases cause T-cell-independent occupational asthma, because asthma-like airway inflammation, including eosinophilia, mucus formation, and AHR, was found in *Rag2*$^{-/-}$ mice, which lack T and B cells.[8] This suggests that Th2 cells are dispensable in the acute phase of allergic lung inflammation. On the other hand, IL-33-deficient lungs failed to show eosinophilic inflammation and mucus production following intranasal administration of a protease allergen,[8] indicating an intrinsic role of IL-33 in AHR induced by protease allergens. IL-33 directly or indirectly activates ILC2s to induce robust production of IL-5 and IL-13. ILC2s are needed during the early phase to provide an interface between

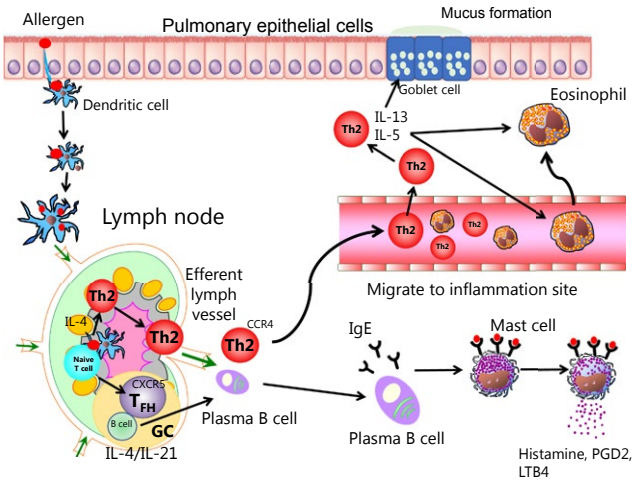


FIGURE 1 Classical model of pulmonary allergic lung inflammation. In atopic asthma, soluble protein allergen is acquired by dendritic cells (DCs) which then migrate into a secondary lymphoid organ, the draining LN, where naive T cells are sensitized with processed and presented allergen peptides. After their activation by DC, naive T cells develop into either adaptive Th2 cells or T_{FH} cells. T_{FH} cells are a specialized T-cell subset that interacts with antigen-specific B cells to induce IgE production. The allergen-specific IgE antibodies bind to the high-affinity FcεR on mast cells. Crosslinking of this IgE by allergen activates mast cells and they release a variety of inflammatory mediators, such as histamine, prostaglandin D2 (PGD2), and leukotriene B4 (LTB4). Th2 cells migrate into the site of inflammation and produce Th2 cytokines after antigen challenge. IL-5 and IL-9 promote tissue eosinophilia and mast cell hyperplasia. IL-13 promotes mucus production by goblet cells and AHR

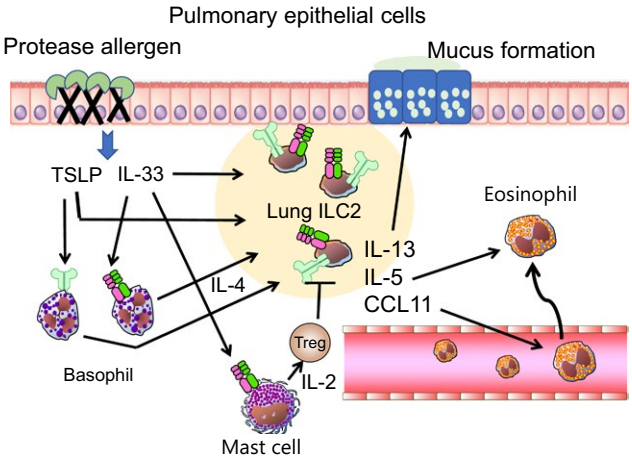


FIGURE 2 Non-atopic lung inflammation. Proteases activity contained in proteases allergens cause barrier disruption in epithelial cells, leading to elevation of IL-33 and TSLP, which activate basophils and ILC2s to induce different Th2 cytokines. The basophil-derived IL-4 synergistically activates ILC2s together with IL-33 to release chemoattractants, such as CCL11, that are required for eosinophilia in the lung. The activated ILC2s also produce IL-5 and IL-13, which cause eosinophilia and mucus production in early airway inflammation. IL-33 also leads to Treg expansion through IL-2 secreted by the activated mast cells

IL-33 and eosinophilic lung inflammation,[9,10] because ILC-deficient mice (*Rag2*$^{-/-}$*Il2rg*$^{-/-}$) had a marked reduction in the characteristic features of lung allergic inflammation, mucus production, and eosinophilia. Therefore, the IL-33-ILC2-IL-5/IL-13 axis is thought to be a critical pathway for protease-induced airway inflammation. Interestingly, basophil-deficient mice failed to show infiltration of eosinophils into the lung or airways, mucus formation, and AHR, suggesting an important contribution of basophils to protease-induced eosinophilic inflammation.[11] It has been recently discovered that innate immune cells directly or indirectly influence Th2 cell-mediated lung inflammation through production of Th2 cytokines.[12] Th2 cytokines secreted from innate immune cells, including ILC2s and eosinophils, have multiple functions, influencing not only the acute phase as direct effector molecules, but also in the chronic phase as an initiator for Th2 polarization as part of their cross-talk with adaptive immunity. This review describes and discusses the role of IL-4 and IL-13 from basophils, ILC2s, and eosinophils in the acute and chronic phases.

2 | IL-33 AND TSLP IN ALLERGIC LUNG INFLAMMATION

IL-33 is a ligand for IL-1RL1 and is expressed not only by immune cells, such as macrophages, dendritic cells (DCs), and mast cells, but also by non-immune cells, such as fibroblasts and endothelial cells, epithelial cells, smooth muscle cells, and synovial cells.[13,14] IL-33 protein is attached to heterochromatin in the nucleus via a helix-turn-helix motif, and tissue damage cause a release of IL-33 from necrotic cells. Thus, IL-33 is defined as an alarmin or a damage-associated

molecular pattern molecule. The IL-33 receptor (IL-33R) complex comprised IL-1RL1 (also called T1/ST2) and IL-1 receptor accessory protein (IL-1RAcP). The IL-33R is a member of the toll/IL-1 receptor superfamily, whose members activate NF-κB and AP-1 through the MyD88-IRAK1/4-TRAF6/MAPK pathway.[14]

Large-scale genome-wide association studies have demonstrated that the *IL33* and *IL1RL1* genes are associated with human asthma.[15,16] IL-33 is highly expressed in asthmatic patients, AD patients, and after anaphylactic shock.[17,18] In a mouse model, IL-33 injection directly induces eosinophilic inflammation and AHR.[19] Interestingly, in *Il1rl1*-deficient mice, there was a clear reduction in AHR and eosinophilic inflammation after short-term sensitization,[18] but no effect on chronic sensitization.[20,21]

TSLP resembles the cytokine IL-7, originally characterized as a family of B-cell-stimulating factors. The receptor for TSLP is a heterodimer comprised of TSLP receptor (TSLPR) and IL-7Rα, and receptor engagement activates STAT5 signaling.[22] Genetic association with *TSLP* has been reported in several allergic diseases, including asthma and eosinophilia, and with serum IgE levels.[23–26] These observations are supported by the high levels of TSLP expression in skin lesions and in sera from AD patients.[27,28] TSLP expression is also known to be a risk factor for food allergies.[29–31] The importance of TSLP in lung inflammation has been demonstrated in that *Tslpr*$^{-/-}$ mice are resistant to AHR responses in a classical antigen sensitization model.[32] Therefore, TSLP promotes allergic inflammation by direct or indirect functions of Th2 cells, basophils, and other granulocyte populations.

3 | MAST CELLS IN ALLERGIC LUNG INFLAMMATION

Mast cells and basophils have specified effector functions in type 2 immunity, originally established in response to parasitic worms (helminths), but also contribute to pro-inflammatory responses to allergens. These two lineages might have a common precursor cell, a granulocyte/monocyte progenitor-type cell, which was responsible for protection from pathogenic infection in primitive chordates. Their functional specification is determined by environment factors and the cytokine milieu in local tissue. Mast cells and basophils express an overlapping set of cytokines and biochemical compounds stored in their cytosolic granules, and many of these compounds are rapidly released by activation of the high-affinity FcεRI with allergen-mediated cross-linking and by other ligand-receptor interactions. The cytosolic granules contain cytokines (IL-4, IL-5, IL-6, and IL-13), biogenic amines (histamine and serotonin), serglycin, proteoglycans, mast cell-derived proteases (chymase and tryptase), and lipid mediators (platelet-activating factor [PAF], leukotrienes, prostaglandins, and sphingolipids). In mouse mast cells, mMCP1, mMCP2, mMCP4, mMCP5, mMCP9, and mMCP10 are specific proteases, and mMCP6 and mMCP7 are specific tryptases.[33,34] On the other hand, in mature basophils, mMCP8 is a specific protease and mMCP11 is a specific tryptase.[35] Degranulation and release of these mast cell- and basophil-associated substances subsequently control the recruitment of inflammatory cells, smooth muscle constriction, and increased vascular permeability.

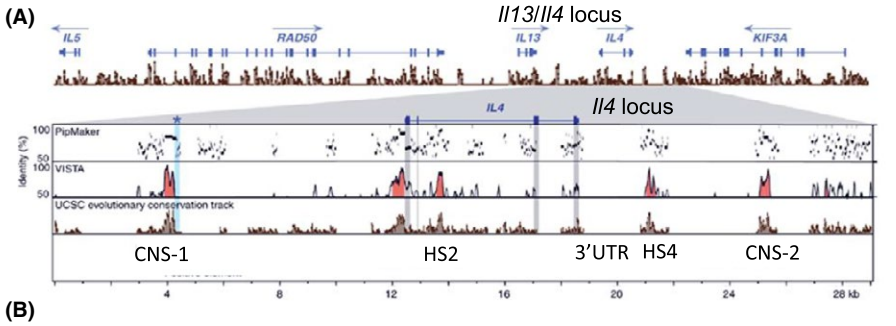
Mast cell activation through aggregation of the FcεRI by cross-linking of IgE with polyvalent antigen is the major driver of allergic inflammation, the passive cutaneous anaphylaxis (PCA) reaction, allergic rhinitis, food allergy, and systemic anaphylaxis (Figure 2). PCA, the classical experimental model for the anaphylactic reaction, is caused by the release of vasoactive substances including histamine as the result of IgE-mediated mast cell activation. Mast cells are also activated by complement components C3a and C5a through binding to their cognate receptors, C3aR and C5aR (CD88), as well as by nerve growth factor. Human mast cells are also activated through TLR3 by double stranded RNA.[36]

Several mutant mice and genetically modified mice have proven useful to address the in vivo function of mast cells.[37–39] These include the Kit-based mast cell-deficient mice, WBB6F1-*Kit*$^{W/W\text{-}v}$ and *Kit*$^{W\text{-}sh/W\text{-}sh}$, and the selective mast cell-deficient mouse models, *Mcpt5*-Cre,[40] *Cpa3*-Cre (Hello *Kit*ty),[41] *Cpa3*Cre,[38] and Mas-TRECK, which uses a diphtheria-toxin receptor (DTR) transgene under the control of the DNase I hypersensitive site 2 (HS2) region of the *Il4* locus (Figure 3). Passive systemic anaphylaxis is well-studied allergic mouse model that has been thought for years to be mediated by mast cells.[42–44] However, recent studies with the mast cell-deficient Mas-TRECK mouse and the alum-OVA asthma model showed AHR responses and eosinophil recruitment comparable to control mice. The relative contribution of mast cells has been demonstrated in some deletion models, using sensitization that omits strong adjuvant effects and that immunizes with low concentrations of antigen. Indeed, reconstitution of *Kit*$^{W\text{-}sh/W\text{-}sh}$ and/or *Kit*$^{W/W\text{-}v}$ mice with mast cells caused lung eosinophilia, tissue remodeling, and enhanced AHR in the absence of adjuvant.[45] Furthermore, DT-mediated conditional depletion of mast cells in Mas-TRECK mice resulted in reduction of AHR responses using a similar sensitization protocol.[39] These observations suggest that mast cells mainly contribute to the IgE-mediated allergic inflammation. This type of allergic lung inflammation may be effectively controlled by a combination of inhaled corticosteroids and a β2-adrenergic agonist, treatments that are routinely used for atopic asthma patients.

Recently, a role for mast cells in regulatory T (Treg) cell biology has been reported in protease allergen- and IL-33-induced lung eosinophilia models.[46] Mast cells activated by a plant-derived protease allergen induced IL-33 and secreted several cytokines, including IL-2. Mast cell deficiency in *Kit*$^{W\text{-}sh/W\text{-}sh}$ mice caused exacerbation in lung inflammation and reduced numbers of Treg cells, suggesting that the mast cell-derived IL-2 suppressed ILC2 function through the expansion of Treg cells. This finding suggests that mast cells have a role in multiple layers of lung allergic inflammation (Figure 2).

4 | BASOPHILS IN ALLERGIC LUNG INFLAMMATION

Basophils are relatively short-lived circulating cells (about 2-3 days) under steady-state conditions and are recruited to inflammatory sites under allergic conditions, while mast cells are long-lived tissue resident cells.[47,48] Runx1 is a transcriptional factor critical for development of basophils, and



IL-4	CNS-1	HS2	3'UTR	HS4	CNS-2
Th2	±	+	−	−	−
T_{FH}	?	+	−	−	+
NKT	?	+	−	−	+
Mast cell	?	+	−	−	+
Basophil	?	−	+	−	+
ILC2	−	−	−	−	−

FIGURE 3 Overview of *cis*-regulatory elements in the *Il13/Il4* locus. (A) The *Il4* locus contains several high homology regions. The bottom column in this figure display homology between human and mouse. These high homology regions significantly overlap with conserved non-coding sequences. The *Il4* locus contains conserved non-cording sequence 1 (CNS1), DNaseI hypersensitive (HS)2, 3′ untranslated region (3′ UTR), HS4, and CNS-2. (B, C) Each enhancer has different activity in different cell lineages. (B) Summary of each *Il4* enhancer based on the results from knockout and transgenic mice. (CGFP expression under the control of the HS2 and 3′ UTR/HS4 enhancers in reporter transgenic mice. HS2 controls IL-4 expression in Th2, T_{FH}, NKT, and mast cells. 3′ UTR controls IL-4 expression in basophils. CNS2 controls IL-4 expression in T_{FH}, NKT, mast cells, and basophils

mice lacking distal promoter-derived Runx1 (P1-Runx1) do not have any basophils, but mast cells are intact.[49] Basophils specifically express FcεRIα and CD49b, basogranulin, and CD203c,[50–52] and have some degree of functional heterogeneity depending on the cytokine environment. It has been reported that basophil lineage effector functions are determined by TSLP, which promotes high levels of IL-18Rα, T1/ST2, IL-4, CCL3, CCL4, CCL12, and CXCL2. TSLP thus controls basophil recruitment to sites of inflammation to promote further Th2 cytokine-mediated inflammation.[53]

The contribution of basophils to the allergic response has been highlighted in IgE-mediated chronic allergic inflammation (IgE-CAI), which is induced by subcutaneous injection of multivalent antigens in mice sensitized with antigen-specific IgE. Delayed-onset ear swelling with massive infiltration of eosinophils is the major pathology seen in IgE-CAI, which is a basophil-dependent allergic response.[54] Basophils also appear to control the anti-inflammatory function of M2 macrophages, which inhibit recruitment of inflammatory monocytes.[55] It has been shown in mouse asthma models that basophil depletion with antibodies against the orphan-activating receptor CD200R3 only partially reduced the number of infiltrated eosinophils into the bronchoalveolar lavage[56] fluid.[57] Constitutive and selective basophil-deficient mouse systems: *Mcpt8*IRES-YFP-Cre, *Mcpt8*-Cre and Bas-TRECK (using a DTR transgene under the control of the HS4 region of the *Il4* locus, see Figure 3), BasoDTR (using a DTR transgene under the control of CD203c promoter) have been useful for exploring in vivo basophil functions.[39,58–60] In two independent deletion systems, Bas-TRECK and *Mcpt8*-Cre mice, the alum-OVA sensitization model showed AHR responses and eosinophil infiltration comparable to control mice.[39,59] In contrast, human studies indicated that basophils have a role in the late phase of allergic asthma.[61] These observations are supported by recent findings in the T-independent protease allergen-mediated acute lung inflammation model. Basophil deletion in Bas-TRECK mice resulted in resolution of the eosinophilia and mucus production induced by nasal administration of a protease allergen.[11]

Importantly, there are some functional differences between human and mouse basophils. In mouse anaphylaxis model, activated basophils profoundly produce the vasoactive lipid mediator PAF, which contribute to the development of IgG1-dependent anaphylaxis.[62] Classical anaphylaxis is known to be promoted by crosslinking of IgE on mast cells, leading to the release of histamine and lipid mediators by degranulation.[63] Studies of $Kit^{W/W-v}$ and $Ige^{-/-}$ mice indicated the presence of mast cell- and IgE-independent pathways for anaphylaxis,[64,65] and treatment with basophil-depleting antibodies implicated the PAF-mediated pathway, activated in response to crosslinking of IgG1 bound to the FcγRIIIA. However, human basophils are not the case and fail to bind to IgG immune complexes.[50,62] Bas-TRECK mice and control mice had comparable anaphylactic responses, suggesting still a questionable role of basophils in such responses (data not shown).

5 | BASOPHIL-DERIVED IL-4 IN ALLERGIC LUNG INFLAMMATION

Basophils express relatively high levels of IL-4 compared to other innate cells including mast cells, ILC2s, and eosinophils.[19,66,67] Therefore,

basophils have been in the spotlight as a major IL-4 source to promote Th2 differentiation. In earlier work, basophil depletion by treatment with the anti-FcεRI antibody (MAR-1) led to the loss of Th2 differentiation.[68,69] Basophils were thought to be important as antigen-presenting cells, because basophils constitutively express detectable level of MHC class II and costimulatory molecules, including CD80, CD86, and CD40.[69,70] However, later work using mice constitutively lacking either basophils or DCs indicated that Th2 cell polarization induced by papain treatment requires DCs but not basophils.[59] Genetic ablation of basophils resulted in normal development of Th2 cells in response to immunization with OVA plus alum adjuvant.[39,58] These findings have called into question the contribution of basophils as antigen-presenting cells in Th2 polarization.

In asthma patients, administration of an antibody against the IL-4 receptor α-chain (dupilumab), which blocks downstream of IL-4 and IL-13 signaling, reduces the frequency of exacerbation in moderate to severe cases and improves lung function.[71] However, in a mouse model of asthma induced by protease allergens, a possible role for IL-4 remained less clear for a long time.[12] On the other hand, in the context of a chronic AD model, basophil-derived IL-4 appears to promote the accumulation of eosinophils in the skin[50,53,72] as well as ILC2-mediated proinflammatory responses in mouse models of TSLP-induced AD and protease allergen-induced asthma.[53,73] Food allergy mouse models have also highlighted a critical role for basophil-derived IL-4.[74–76] It has been reported that the accumulation of TSLP-elicited basophils promotes epicutaneous Th2 sensitization to food antigens and is critical for the development of IgE-mediated food allergy.[77] TSLP-elicited basophils rather than Th2 cells are the dominant source of IL-4 in inflamed AD skin.[78] A number of mouse studies have demonstrated that skin barrier defects contribute to the emergence of allergic inflammation at the epithelial barrier through epicutaneous antigen sensitization.[79,80] In this context, antigen sensitization at an AD-like skin lesion eventually leads to IgE-mediated intestinal food allergy.[77]

To understand the role of IL-4 derived from innate and adaptive immune cells, several regulatory elements were characterized in the *Il13*/*Il4* locus (Figure 3); conserved sequence 1 (HSS1 and HSS2), located between *Il13* and *Il4*, HS0, located in the 5′ region of *Il4*, HS1, located in the promoter region, HS2 and HS3 in the second exon, and HS5a and HS5, located 3′ of *Il4*.[81–83] HS2 in the *Il4* locus is a critical enhancer that regulates Th2- and mast cell-specific *Il4* expression.[84,85] The HS5a and HS5 region, also called conserved non-coding sequence 2, is an essential enhancer for IL-4 expression in mast cells and T follicular helper (T_{FH}) cells, but not in Th2 cells.[86,87] IL-4 expression in basophils is controlled by distinct enhancers, the 3′ UTR is a unique enhancer controlling basophil-specific IL-4. This series of studies indicated that IL-4 expression is regulated by distinct enhancers in mast cells, basophils and Th2 cells, and this distinct regulatory arrangement provides us a useful way to investigate a role of IL-4 in basophil-specific IL-4-deficient mice and to establish the DT-mediated in vivo mast cell and basophil depletion system, Mas-TRECK and Bas-TRECK.

There was a reduction of protease allergen-induced lung inflammation with the loss of basophil-derived IL-4, as shown in mice lacking the *Il4* 3′ UTR and by reconstitution of basophil-deficient mice with *Il4*-deficient basophils. Therefore, basophil-derived IL-4 has a critical role for ILC2 activation in acute papain challenge (Figure 2).[11] However, ILC2s also secrete IL-4 in a particular case of in vivo antigen challenge, which raises the possibility that ILC2-derived IL-4 might act as an autocrine factor.[88] The importance of basophil-derived IL-4 was also demonstrated in allergic skin reactions in a mouse model. Skin administration of a vitamin D analog (MC903) induced epithelial cell-derived TSLP that initiated basophil activation and accumulation of ILC2s, which contribute to Th2 cytokine-mediated AD-like skin lesions.[35,49] The basophil-derived IL-4 was required for the optimal accumulation of ILC2s in the inflamed skin lesions[78] and for epicutaneous sensitization to food antigens in the skin and in food allergy effector responses.[89] Indeed, mice lacking the *Il4* 3′ UTR fail to develop IgE-mediated food allergy.

6 | ILC2 IN ALLERGIC INFLAMMATION

ILC2 were originally discovered in the gut-associated mucosal tissues.[66,90,91] They were identified in different laboratories and originally had several designations—innate helper type 2 (Ih2) cells, multipotent progenitor type 2 (MPP^{type2}) cells, natural helper cells, and nuocytes—but are now classified as ILC2s.[92] ILC2s produce high levels of IL-5, IL-6, and IL-13 in response to epithelial or myeloid cell-derived IL-25, IL-33, and TSLP after exposure to protease allergens, chemicals, or helminth parasites.[9,67,89,93–97] These innate lymphocytes have also been found in several other tissues including the liver, spleen, and lung.[67] The importance of ILC2s cells has been reported in viral-induced airway inflammation in BALB/c mice.[98] Furthermore, it has been suggested that IL-13-producing ILCs may play a role in lung eosinophilic inflammation and asthma.[2,99]

The functional importance of ILC2s in type 2 immunity has been demonstrated using reconstitution with ILC2 cells in gene targeted mice, genetically deleted mice, or temporarily ablated mice.[19,66] Expansion of ILC2s has been shown not only in asthmatic inflammation observed in both human and mouse,[9,56,93,98,100] but also in the nasal polyps of human patients with chronic rhinosinusitis.[94] ILC2s have been also found circulating in the blood of asthma patients[101] and in the BALF from idiopathic pulmonary fibrosis patients.[102] ILC2 commonly express CD90.2 (Thy1), CD127 (IL7Rα), KLRG1, and ICOS, T1/ST2 (IL33R), and CD25 (IL-2Rα), and contain *Il13*, *Il5*, *Il4*, *Il9r*, and *Il17rb* transcripts. These varied among tissues, but show quite characteristic signatures of transcriptome in comparison with ILC1 and ILC3.[103,104] Early studies demonstrated that IL-2 and IL-7 potently induce proliferation of ILC2s and that signaling through the common γ (γc) receptor (also called IL-2Rγ) is critical for ILC2 development.[66,67,90] More complex patterns of γc-ligands, IL-4 and IL-9, have been identified as regulators of ILC2 function and activation.[11,105] Recently, higher expression of KLRG1 by ILC2s has been reported to dictate inflammatory function in the response to IL-25, thus $KLRG1^{high}$ ILCs are potentially the inflammatory subset.[106] Therefore, cytokine-dependent expansion of effector-type ILC2s may drive a chronic inflammatory situation in the lung.

However, the in vivo contributions of ILC2 cells in allergic lung inflammation still remain unclear. ILC2-deficient mice (CD45 congenic chimeric mice transferred with bone marrow from RAR-related orphan receptor alpha [Rora$^{sg/sg}$] mice) exhibited less-severe lung inflammation and about one-third the levels of eosinophil migration and IL-13 production in comparison with ILC2-sufficient mice. The reduction was only seen in the response to intranasal administration of protease allergens, such as papain and HDMs, but not systemically to the Th2-inducing antigen OVA.[107] Moreover, the process of airway remodeling was impaired by ILC2 depletion, but not by CD4$^+$ T-cell depletion, in an experimental mouse model of chronic asthma, suggesting that ILC2 cells contribute to airway remodeling.[108] ILC2 cells constitutively express the epidermal growth factor family member amphiregulin, which is a key molecule to control airway remodeling by acting on repair processes of fibroblasts and epithelial cells.[93,109] Moreover, ILC2 cells have been shown to control macrophage activation, which promotes airway wall remodeling through collagen synthesis.[110] Therefore, ILC2 cells are also required for tissue repair in the chronic phase of allergic lung inflammation.

7 | TH2 POLARIZATION IN ALLERGIC INFLAMMATION AND THE DC CONTRIBUTION

The exposure to allergens causes Th2 cell activation and secretion of various type 2 cytokines including IL-4. IL-4 was originally identified as B-cell-stimulating factor,[111] and induces germline γ1 and ε transcripts, subsequently leading to class switch recombination and production of IgG1 and IgE,[112–114] and antigen cross-linking of the IgE on mast cells and basophils is strongly implicated in atopic and allergic disease. Classically, T_H2-derived IL-4 has been thought to be responsible for IgE responses, but T_{FH} cells are currently considered an alternative IL-4 source to mainly regulate IgE responses to soluble antigen and to parasitic infection (Figure 1).[86,87] It had been thought that IL-4 was critical for Th2 cell differentiation, which is controlled by the expression of the key transcription factor GATA3.[85] However, the driving signal for Th2 polarization has been a long-standing question. Indeed, the initial IL-4 source responsible for the Th2 differentiation has been undefined, although there are several candidates, including naive CD4 T cells,[115] memory phenotype T cells,[116] natural killer T cells, basophils,[11,84] and DCs.

Accumulating recent evidence has suggested that a CD11b$^+$ DC subset might trigger Th2 differentiation. DT-mediated depletion of CD11c$^+$ cells in DTR transgenic mice under control of the CD11c/*Itgax* promoter markedly impairs Th2 responses,[117] and CD11b$^+$ DC cells expressing FcεR1 are responsible for in vivo type 2 immune responses in the response to HDM.[57] These CD11b$^+$ conventional DCs (cDCs) are the only DC subset able to migrate into the LN to trigger Th2 responses.[118] On the other hand, both CD11b$^+$ cDCs and monocyte-derived DCs (moDCs) induced recruitment of Th2 cells to lung airways in the response to HDM. The moDC is a lung residential DC subset able to induce Th2 cell-mediated AHR only at a high dose of HDM. In contrast, under conditions of physiological low-dose exposure to HDM, CD11b$^+$ cDCs have the exclusive function to induce Th2 cell-mediated AHR, because *Flt3l*$^{-/-}$ mice lacking migratory and resident cDCs showed no eosinophilic airway inflammation.[119]

Recent studies indicated involvement of the IRF4$^+$ cDC in Th2 responses induced by HDM (Figure 4).[120] Two other studies argued that CD301b$^+$ and/or PD-L2$^+$ dermal DCs control IRF4-dependent Th2 responses,[121,122] although the *Irf4*$^{-/-}$ mice showed an impairment in DC migration from local tissues to LNs.[123] IRF4 is known to control the expression of MHC class II antigen and costimulatory molecules, which are required for tight interaction between antigen presentation and T cells.[124] However, IRF4 was dispensable for the in vivo expression of MHC II,[123] indicating that the defect in DC migration is not related to class II expression. A subset of CD11b$^-$ IRF4$^+$ cDCs has been reported to be essential for Th2 responses, and the generation of this subset in the skin draining LNs is dependent on the transcription factor KLF4.[125] Involvement of CD326^{lo}CD103^{lo}CD11b^{lo} dermal DCs has also been suggested in Th2 responses during contact sensitization in mice.[126]

IRF4 is also necessary for development of CD11b$^+$CD172 (SIRP1α)$^+$ cDCs, which are sufficient to induce allergic sensitization.[127] Moreover, *Klf4* deficiency caused a reduction in the number of CD11b$^+$ cDCs in the spleen,[128] and CD11c-specific *Klf4* conditional knockout mice had a reduction of Th2 cell responses after HDM challenge and selective loss of IRF4 expressing cDC.[125]

8 | AMPLIFICATION OF TH2 IMMUNITY BY CELLULAR ORCHESTRATION VIA CYTOKINES

ILC2 cells directly or indirectly influence Th2 cell-mediated lung inflammation through the Th2 cytokines. Although, based on in vitro evidence, the Th2 differentiation process is mainly controlled by IL-4, several recent studies have suggested the importance of IL-13 for Th2 cell-mediated lung inflammation in the initial response to protease allergens such as papain and HDM. ILC2 cells may be an early source of IL-13 required for polarization of naive CD4$^+$ T cells into Th2 cells. Indeed, it has been reported that ILC2-derived IL-13 is critical to promote the migration of CD11b$^+$ cDCs into the draining LN where they drive naive T cells to become Th2 cells (Figure 4).[12] Thus, this migration is important and sufficient for Th2 development. The IL-13 derived from ILC2s also induces CCL17 production from IRF4$^+$CD11b$^+$ DCs to attract Th2 cells.[129,130] However, how IL-13 controls the migratory function of cDCs still remains to be defined.

Interestingly, ILC2 and ILC3 cells seem to express MHC class II and ICOS.[66,131] The expression level of MHC class II was found at a low level in the steady state, but the expression increased during the sensitization phase. MHC expression was required for efficient Th2 cell generation during *Nippostrongylus brasiliensis* infection,[88] leading to the hypothesis that ILC2 cells may act as antigen-presenting cells. ILC2 cells present OVA peptide and induce proliferation of OVA-specific T cells in OTII transgenic (OTII Tg) mice when antigenic peptides are present in the culture. Furthermore, human ILC2 cells could drive the

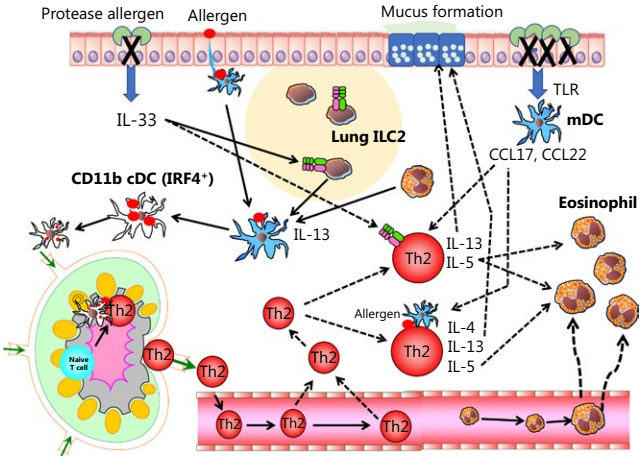


FIGURE 4 Innate and adaptive immunity crosstalk during the sensitization and challenge phase of lung inflammation. In the sensitization phase (solid lines), pulmonary epithelial cells can be activated directly by protease allergens. In response, lung epithelial cells release IL-33 that directly or indirectly activates ILC2 to produce IL-13. In some cases, lung eosinophils could be an alternative source of IL-13. IL-13 drives the maturation of CD11b+ cDCs, which depend on the transcription factor IRF4 for their maturation from pre-cDCs. After activation, the $CD11b^+$ cDCs migrate into the draining mediastinal lymph nodes, where naive T cells differentiate into Th2 cells. During the challenge phase (dashed lines), DCs again have a predominant role in Th2 effector function. A high dose of protease allergen activates monocytic DCs (moDCs) and recruits effector Th2 cells through the production of CCL17 and CCL22. In this case, moDCs need to express pattern-recognition receptors, such as TLR, to stimulate release of these chemokines. The pulmonary epithelial cell-derived IL-33 directly activates effector Th2 cells to produce IL-5 and IL-13, while the allergen-acquired DCs activated the production of all Th2 cytokines, including IL-4, IL-5, and IL-13, in the response to allergen

ex vivo proliferation of $CD4^+$ T cells specific for HDM.[88] However, a recent report indicated that loss of MHC class II expression on ILC2s made no difference in the number of lung ILC2s, eosinophils, or IL-5-expressing effector Th2 cells.[132] Therefore, whether ILC2 cells are necessary for antigen presentation and, if so, how ILC2 cells can migrate into the draining LN in vivo both remain to be addressed.

Re-exposure to the same allergens induces a robust activation of memory Th2 cells or long-lived Th2 cells in the draining LN. The memory Th2 cells acquire high levels of T1/ST-2 expression, and epithelial cell-derived IL-33 promotes Th2 activation in the pathogenesis of allergic airway inflammation in both mouse and human. The activated memory OTII Th2 cells secreted large amounts of Th2 cytokines in response to OVA, and IL-5 production only depended on IL-33.[133] Allergen non-specific Th2 cells also make critical contributions to the cysteine protease-induced eosinophilic inflammation as a result of re-localizing from the draining LN into the inflammatory site. These Th2 cells can express high levels of IL-5 and IL-13 in response to IL-33 but independent of antigen recognition, which is generally required for initial T-cell activation.[134] Targeted depletion of ILC2 cells profoundly impairs Th2 cell localization to the lungs after allergen re-challenge, suggesting that ILC2 cells are crucial for Th2 cell-mediated responses (Figure 4). However, it was also reported that substantial deletion of ILC2s in a Rag-deficient background with T-cell reconstitution had no effect on activation of adaptive Th2 cell responses during infection with *N. brasiliensis*.[132]

These findings also suggest that ILC2s are not the only source of IL-13 required for DC licensing in the activation of Th2 cells. Involvement of other cell types remains to be further defined. A role for eosinophils was reported in DC regulation to promote the Th2 polarization characteristic of the pulmonary microenvironment in asthmatic responses (Figure 4).[135] Experimental reconstitution of eosinophil-deficient *PHIL* mice with eosinophils from *Il4*$^{-/-}$ or *Il13*$^{-/-}$ mice indicated a critical role of eosinophil IL-13, but not of IL-4, in Th2 cell-mediated lung inflammation.[136] On the other hand, mast cell-derived IL-13 also has a role to increase the frequency of PD-$L2^+$ cDCs that control Th2 cell-mediated responses in the lung after a single dose of intranasal *Aspergillus fumigatus* protease (M. Cho, H. Lim, Y. Chung, unpublished data). Further investigations of the link between innate and adaptive responses will be beneficial to our understanding of the complexity of asthmatic pathology and to improve treatments based on knowledge of different disease mechanisms in individual asthma patients.

9 | CLOSING REMARKS

IL-13 secreted from innate immune cells, including ILC2s and eosinophils, promotes the migration of $IRF4^+CD11b^+$ DCs into the draining LN in the acute phase, and the $CD11b^+$ DCs then initiate Th2 polarization. However, major questions remain regarding how IL-13 controls the phenotypic change and the migratory function of the cDCs. There is no clear understanding of the molecular mechanisms underlying Th2 polarization controlled by the $IRF4^+CD11b^+$ DCs in the LN or how the polarized Th2 cells then migrate back to the pulmonary allergic inflammatory site. Although IL-33-mediated Th2 activation is clearly occurring, its signaling cascade remains unclear, and it still unclear why IL-33 selectively controls IL-5 and IL-13, but not IL-4 expression, in the primed Th2 cells. Moreover, if IL-4 is dispensable in the chronic phase of Th2-mediated pulmonary allergic inflammation, what is the main role of IL-4? It will also be important to determine if DC subsets corresponding to DCs in the mouse exist in humans. Answering these questions is important to reevaluate therapeutic strategies and targets and to develop new ones.

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CONFLICT OF INTEREST

There is no conflict of interest.

REFERENCES

1. Kim HY, DeKruyff RH, Umetsu DT. The many paths to asthma: phenotype shaped by innate and adaptive immunity. *Nat Immunol*. 2010;11:577-584.
2. Lloyd CM, Hessel EM. Functions of T cells in asthma: more than just T(H)2 cells. *Nat Rev Immunol*. 2010;10:838-848.
3. Paul WE. Endless fascination. *Annu Rev Immunol*. 2014;32:1-24.
4. Pulendran B, Artis D. New paradigms in type 2 immunity. *Science*. 2012;337:431-435.
5. Zhu J, Yamane H, Paul WE. Differentiation of effector CD4 T cell populations. *Annu Rev Immunol*. 2010;28:445-489.
6. Gregory LG, Lloyd CM. Orchestrating house dust mite-associated allergy in the lung. *Trends Immunol*. 2011;32:402-411.
7. Novey HS, Marchioli LE, Sokol WN, Wells ID. Papain-induced asthma-physiological and immunological features. *J Allergy Clin Immunol*. 1979;63:98-103.
8. Oboki K, Ohno T, Kajiwara N, et al. IL-33 is a crucial amplifier of innate rather than acquired immunity. *Proc Natl Acad Sci USA*. 2010;107:18581-18586.
9. 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.
10. Kondo Y, Yoshimoto T, Yasuda K, et al. Administration of IL-33 induces airway hyperresponsiveness and goblet cell hyperplasia in the lungs in the absence of adaptive immune system. *Int Immunol*. 2008;20:791-800.
11. Motomura Y, Morita H, Moro K, et al. Basophil-derived interleukin-4 controls the function of natural helper cells, a member of ILC2s, in lung inflammation. *Immunity*. 2014;40:758-771.
12. Halim TY, Steer CA, Matha L, et al. Group 2 innate lymphoid cells are critical for the initiation of adaptive T helper 2 cell-mediated allergic lung inflammation. *Immunity*. 2014;40:425-435.
13. Nakae S, Morita H, Ohno T, Arae K, Matsumoto K, Saito H. Role of interleukin-33 in innate-type immune cells in allergy. *Allergol Int*. 2013;62:13-20.
14. Liew FY, Pitman NI, McInnes IB. Disease-associated functions of IL-33: the new kid in the IL-1 family. *Nat Rev Immunol*. 2010;10:103-110.
15. Grotenboer NS, Ketelaar ME, Koppelman GH, Nawijn MC. Decoding asthma: translating genetic variation in IL33 and IL1RL1 into disease pathophysiology. *J Allergy Clin Immunol*. 2013;131:856-865.
16. Hirota T, Takahashi A, Kubo M, et al. Genome-wide association study identifies three new susceptibility loci for adult asthma in the Japanese population. *Nat Genet*. 2011;43:893-896.
17. Prefontaine D, Lajoie-Kadoch S, Foley S, et al. Increased expression of IL-33 in severe asthma: evidence of expression by airway smooth muscle cells. *J Immunol*. 2009;183:5094-5103.
18. Kurowska-Stolarska M, Stolarski B, Kewin P, et al. IL-33 amplifies the polarization of alternatively activated macrophages that contribute to airway inflammation. *J Immunol*. 2009;183:6469-6477.
19. Liang HE, Reinhardt RL, Bando JK, Sullivan BM, Ho IC, Locksley RM. Divergent expression patterns of IL-4 and IL-13 define unique functions in allergic immunity. *Nat Immunol*. 2011;13:58-66.
20. Hoshino K, Kashiwamura S, Kuribayashi K, et al. The absence of interleukin 1 receptor-related T1/ST2 does not affect T helper cell type 2 development and its effector function. *J Exp Med*. 1999;190:1541-1548.
21. Mangan NE, Dasvarma A, McKenzie AN, Fallon PG. T1/ST2 expression on Th2 cells negatively regulates allergic pulmonary inflammation. *Eur J Immunol*. 2007;37:1302-1312.
22. Ziegler SF, Artis D. Sensing the outside world: TSLP regulates barrier immunity. *Nat Immunol*. 2010;11:289-293.
23. Gudbjartsson DF, Bjornsdottir US, Halapi E, et al. Sequence variants affecting eosinophil numbers associate with asthma and myocardial infarction. *Nat Genet*. 2009;41:342-347.
24. He JQ, Hallstrand TS, Knight D, et al. A thymic stromal lymphopoietin gene variant is associated with asthma and airway hyperresponsiveness. *J Allergy Clin Immunol*. 2009;124:222-229.
25. Gao PS, Rafaels NM, Mu D, et al. Genetic variants in thymic stromal lymphopoietin are associated with atopic dermatitis and eczema herpeticum. *J Allergy Clin Immunol*. 2010;125(1403–1407):e1404.
26. Margolis DJ, Kim B, Apter AJ, et al. Thymic stromal lymphopoietin variation, filaggrin loss of function, and the persistence of atopic dermatitis. *JAMA Dermatol*. 2014;150:254-259.
27. Soumelis V, Reche PA, Kanzler H, et al. Human epithelial cells trigger dendritic cell mediated allergic inflammation by producing TSLP. *Nat Immunol*. 2002;3:673-680.
28. Lee EB, Kim KW, Hong JY, Jee HM, Sohn MH, Kim KE. Increased serum thymic stromal lymphopoietin in children with atopic dermatitis. *Pediatr Allergy Immunol*. 2010;21:e457-e460.
29. Lack G, Fox D, Northstone K, Golding J; Avon Longitudinal Study of Parents and Children Study Team. Factors associated with the development of peanut allergy in childhood. *N Engl J Med*. 2003;348:977-985.
30. Tsakok T, Marrs T, Mohsin M, et al. Does atopic dermatitis cause food allergy? A systematic review. *J Allergy Clin Immunol*. 2016;137:1071-1078.
31. Brough HA, Liu AH, Sicherer S, et al. Atopic dermatitis increases the effect of exposure to peanut antigen in dust on peanut sensitization and likely peanut allergy. *J Allergy Clin Immunol*. 2015;135:164-170.
32. Al-Shami A, Spolski R, Kelly J, Keane-Myers A, Leonard WJ. A role for TSLP in the development of inflammation in an asthma model. *J Exp Med*. 2005;202:829-839.
33. Gallwitz M, Reimer JM, Hellman L. Expansion of the mast cell chymase locus over the past 200 million years of mammalian evolution. *Immunogenetics*. 2006;58:655-669.
34. Pejler G, Ronnberg E, Waern I, Wernersson S. Mast cell proteases: multifaceted regulators of inflammatory disease. *Blood*. 2010;115:4981-4990.
35. Ugajin T, Kojima T, Mukai K, et al. Basophils preferentially express mouse Mast Cell Protease 11 among the mast cell tryptase family in contrast to mast cells. *J Leukoc Biol*. 2009;86:1417-1425.
36. Galli SJ. Mast cells and basophils. *Curr Opin Hematol*. 2000;7:32-39.
37. Dudeck A, Dudeck J, Scholten J, et al. Mast cells are key promoters of contact allergy that mediate the adjuvant effects of haptens. *Immunity*. 2011;34:973-984.
38. Feyerabend TB, Weiser A, Tiets A, et al. Cre-mediated cell ablation contests mast cell contribution in models of antibody- and T cell-mediated autoimmunity. *Immunity*. 2011;35:832-844.
39. Sawaguchi M, Tanaka S, Nakatani Y, et al. Role of mast cells and basophils in IgE responses and in allergic airway hyperresponsiveness. *J Immunol*. 2012;188:1809-1818.
40. Scholten J, Hartmann K, Gerbaulet A, et al. Mast cell-specific Cre/loxP-mediated recombination in vivo. *Transgenic Res*. 2008;17:307-315.
41. Lilla JN, Chen CC, Mukai K, et al. Reduced mast cell and basophil numbers and function in Cpa3-Cre; Mcl-1 fl/fl mice. *Blood*. 2011;118:6930-6938.
42. Park SJ, Cho SS, Oh SS, et al. Promoted expression of mast cell-specific proteases in IgE-dependent passive cutaneous anaphylaxis responses. *Clin Chim Acta*. 2001;314:231-236.
43. Thomas WR, Schrader JW. Delayed hypersensitivity in mast-cell-deficient mice. *J Immunol*. 1983;130:2565-2567.
44. Zhou JS, Xing W, Friend DS, Austen KF, Katz HR. Mast cell deficiency in Kit(W-sh) mice does not impair antibody-mediated arthritis. *J Exp Med*. 2007;204:2797-2802.
45. Yu M, Tsai M, Tam SY, Jones C, Zehnder J, Galli SJ. Mast cells can promote the development of multiple features of chronic asthma in mice. *J Clin Invest*. 2006;116:1633-1641.

46. Morita H, Arae K, Unno H, et al. An interleukin-33-mast cell-interleukin-2 axis suppresses papain-induced allergic inflammation by promoting regulatory T cell numbers. *Immunity*. 2015;43:175-186.
47. Ohnmacht C, Voehringer D. Basophil effector function and homeostasis during helminth infection. *Blood*. 2009;113:2816-2825.
48. Kiernan JA. Production and life span of cutaneous mast cells in young rats. *J Anat*. 1979;128:225-238.
49. Mukai K, BenBarak MJ, Tachibana M, et al. Critical role of P1-Runx1 in mouse basophil development. *Blood*. 2012;120:76-85.
50. Siracusa MC, Kim BS, Spergel JM, Artis D. Basophils and allergic inflammation. *J Allergy Clin Immunol*. 2013;132:789-801; quiz 788.
51. McEuen AR, Buckley MG, Compton SJ, Walls AF. Development and characterization of a monoclonal antibody specific for human basophils and the identification of a unique secretory product of basophil activation. *Lab Invest*. 1999;79:27-38.
52. Buhring HJ, Streble A, Valent P. The basophil-specific ectoenzyme E-NPP3 (CD203c) as a marker for cell activation and allergy diagnosis. *Int Arch Allergy Immunol*. 2004;133:317-329.
53. Siracusa MC, Saenz SA, Hill DA, et al. TSLP promotes interleukin-3-independent basophil haematopoiesis and type 2 inflammation. *Nature*. 2011;477:229-233.
54. Mukai K, Matsuoka K, Taya C, et al. Basophils play a critical role in the development of IgE-mediated chronic allergic inflammation independently of T cells and mast cells. *Immunity*. 2005;23:191-202.
55. Egawa M, Mukai K, Yoshikawa S, et al. Inflammatory monocytes recruited to allergic skin acquire an anti-inflammatory M2 phenotype via basophil-derived interleukin-4. *Immunity*. 2013;38:570-580.
56. Barlow JL, Flynn RJ, Ballantyne SJ, McKenzie AN. Reciprocal expression of IL-25 and IL-17A is important for allergic airways hyperreactivity. *Clin Exp Allergy*. 2011;41:1447-1455.
57. Hammad H, Plantinga M, Deswarte K, et al. Inflammatory dendritic cells–not basophils–are necessary and sufficient for induction of Th2 immunity to inhaled house dust mite allergen. *J Exp Med*. 2010;207:2097-2111.
58. Sullivan BM, Liang HE, Bando JK, et al. Genetic analysis of basophil function in vivo. *Nat Immunol*. 2011;12:527-535.
59. Ohnmacht C, Schwartz C, Panzer M, Schiedewitz I, Naumann R, Voehringer D. Basophils orchestrate chronic allergic dermatitis and protective immunity against helminths. *Immunity*. 2010;33:364-374.
60. Matsuoka K, Shitara H, Taya C, Kohno K, Kikkawa Y, Yonekawa H. Novel basophil- or eosinophil-depleted mouse models for functional analyses of allergic inflammation. *PLoS ONE*. 2013;8:e60958.
61. Lichtenstein LM, Bochner BS. The role of basophils in asthma. *Ann N Y Acad Sci*. 1991;629:48-61.
62. Tsujimura Y, Obata K, Mukai K, et al. Basophils play a pivotal role in immunoglobulin-G-mediated but not immunoglobulin-E-mediated systemic anaphylaxis. *Immunity*. 2008;28:581-589.
63. Simons FE. Anaphylaxis. *J Allergy Clin. Immunol*. 2010;125:S161-S181.
64. Oettgen HC, Martin TR, Wynshaw-Boris A, Deng C, Drazen JM, Leder P. Active anaphylaxis in IgE-deficient mice. *Nature*. 1994;370:367-370.
65. Jacoby W, Cammarata PV, Findlay S, Pincus SH. Anaphylaxis in mast cell-deficient mice. *J Invest Dermatol*. 1984;83:302-304.
66. Neill DR, Wong SH, Bellosi A, et al. Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity. *Nature*. 2010;464:1367-1370.
67. Price AE, Liang HE, Sullivan BM, et al. Systemically dispersed innate IL-13-expressing cells in type 2 immunity. *Proc Natl Acad Sci USA*. 2010;107:11489-11494.
68. Sokol CL, Barton GM, Farr AG, Medzhitov R. A mechanism for the initiation of allergen-induced T helper type 2 responses. *Nat Immunol*. 2008;9:310-318.
69. Perrigoue JG, Saenz SA, Siracusa MC, et al. MHC class II-dependent basophil-CD4+ T cell interactions promote T(H)2 cytokine-dependent immunity. *Nat Immunol*. 2009;10:697-705.
70. Yoshimoto T, Yasuda K, Tanaka H, et al. Basophils contribute to T(H)2-IgE responses in vivo via IL-4 production and presentation of peptide-MHC class II complexes to CD4+ T cells. *Nat Immunol*. 2009;10:706-712.
71. Wenzel S, Ford L, Pearlman D, et al. Dupilumab in persistent asthma with elevated eosinophil levels. *N Engl J Med*. 2013;368:2455-2466.
72. Obata K, Mukai K, Tsujimura Y, et al. Basophils are essential initiators of a novel type of chronic allergic inflammation. *Blood*. 2007;110:913-920.
73. Kim BS, Siracusa MC, Saenz SA, et al. TSLP elicits IL-33-independent innate lymphoid cell responses to promote skin inflammation. *Sci Transl Med*. 2013;5:170ra116.
74. Dourado LP, Saldanha JC, Gargiulo DL, et al. Role of IL-4 in aversion induced by food allergy in mice. *Cell Immunol*. 2010;262:62-68.
75. Cardoso CR, Provinciatto PR, Godoi DF, et al. IL-4 regulates susceptibility to intestinal inflammation in murine food allergy. *Am J Physiol Gastrointest Liver Physiol*. 2009;296:G593-G600.
76. Burton OT, Darling AR, Zhou JS, et al. Direct effects of IL-4 on mast cells drive their intestinal expansion and increase susceptibility to anaphylaxis in a murine model of food allergy. *Mucosal Immunol*. 2013;6:740-750.
77. Noti M, Kim BS, Siracusa MC, et al. Exposure to food allergens through inflamed skin promotes intestinal food allergy through the thymic stromal lymphopoietin-basophil axis. *J Allergy Clin Immunol*. 2014;133:1390-1399, 1399 e1391-1396.
78. Kim BS, Wang K, Siracusa MC, et al. Basophils promote innate lymphoid cell responses in inflamed skin. *J Immunol*. 2014;193:3717-3725.
79. Han H, Xu W, Headley MB, et al. Thymic stromal lymphopoietin (TSLP)-mediated dermal inflammation aggravates experimental asthma. *Mucosal Immunol*. 2012;5:342-351.
80. Bartnikas LM, Gurish MF, Burton OT, et al. Epicutaneous sensitization results in IgE-dependent intestinal mast cell expansion and food-induced anaphylaxis. *J Allergy Clin Immunol*. 2013;131:451-460):e451–e456.
81. Agarwal S, Rao A. Modulation of chromatin structure regulates cytokine gene expression during T cell differentiation. *Immunity*. 1998;9:765-775.
82. Agarwal S, Avni O, Rao A. Cell-type-restricted binding of the transcription factor NFAT to a distal IL-4 enhancer in vivo. *Immunity*. 2000;12:643-652.
83. Lee DU, Agarwal S, Rao A. Th2 lineage commitment and efficient IL-4 production involves extended demethylation of the IL-4 gene. *Immunity*. 2002;16:649-660.
84. Yagi R, Tanaka S, Motomura Y, Kubo M. Regulation of the Il4 gene is independently controlled by proximal and distal 3' enhancers in mast cells and basophils. *Mol Cell Biol*. 2007;27:8087-8097.
85. Tanaka S, Motomura Y, Suzuki Y, et al. The enhancer HS2 critically regulates GATA-3-mediated Il4 transcription in T(H)2 cells. *Nat Immunol*. 2011;12:77-85.
86. Harada Y, Tanaka S, Motomura Y, et al. The 3' enhancer CNS2 is a critical regulator of interleukin-4-mediated humoral immunity in follicular helper T cells. *Immunity*. 2012;36:188-200.
87. Vijayanand P, Seumois G, Simpson LJ, et al. Interleukin-4 production by follicular helper T cells requires the conserved Il4 enhancer hypersensitivity site V. *Immunity*. 2012;36:175-187.
88. Oliphant CJ, Hwang YY, Walker JA, et al. MHCII-mediated dialog between group 2 innate lymphoid cells and CD4(+) T cells potentiates type 2 immunity and promotes parasitic helminth expulsion. *Immunity*. 2014;41:283-295.
89. Hussain M, Borcard L, Noti M, et al. Basophil-derived interleukin-4 promotes epicutaneous antigen sensitization concomitant with the development of food allergy. *J Allergy Clin Immunol*. 2017; pii: S0091-6749(17)30566-3.
90. Moro K, Yamada T, Tanabe M, et al. Innate production of T(H)2 cytokines by adipose tissue-associated c-Kit(+)Sca-1(+) lymphoid cells. *Nature*. 2010;463:540-544.

91. Saenz SA, Siracusa MC, Perrigoue JG, et al. IL25 elicits a multipotent progenitor cell population that promotes T(H)2 cytokine responses. *Nature*. 2010;464:1362-1366.
92. Spits H, Artis D, Colonna M, et al. Innate lymphoid cells–a proposal for uniform nomenclature. *Nat Rev Immunol*. 2013;13:145-149.
93. Monticelli LA, Sonnenberg GF, Abt MC, et al. Innate lymphoid cells promote lung-tissue homeostasis after infection with influenza virus. *Nat Immunol*. 2011;12:1045-1054.
94. Mjosberg JM, Trifari S, Crellin NK, et al. Human IL-25- and IL-33-responsive type 2 innate lymphoid cells are defined by expression of CRTH2 and CD161. *Nat Immunol*. 2011;12:1055-1062.
95. Salimi M, Barlow JL, Saunders SP, et al. A role for IL-25 and IL-33-driven type-2 innate lymphoid cells in atopic dermatitis. *J Exp Med*. 2013;210:2939-2950.
96. Bartemes KR, Iijima K, Kobayashi T, Kephart GM, McKenzie AN, Kita H. IL-33-responsive lineage- CD25+ CD44(hi) lymphoid cells mediate innate type 2 immunity and allergic inflammation in the lungs. *J Immunol*. 2012;188:1503-1513.
97. Kabata H, Moro K, Fukunaga K, et al. Thymic stromal lymphopoietin induces corticosteroid resistance in natural helper cells during airway inflammation. *Nat Commun*. 2013;4:2675.
98. Chang YJ, Kim HY, Albacker LA, et al. Innate lymphoid cells mediate influenza-induced airway hyper-reactivity independently of adaptive immunity. *Nat Immunol*. 2011;12:631-638.
99. Kaiko GE, Foster PS. New insights into the generation of Th2 immunity and potential therapeutic targets for the treatment of asthma. *Curr Opin Allergy Clin Immunol*. 2011;11:39-45.
100. Klein Wolterink RG, Kleinjan A, Van Nimwegen M, et al. Pulmonary innate lymphoid cells are major producers of IL-5 and IL-13 in murine models of allergic asthma. *Eur J Immunol*. 2012;42:1106-1116.
101. Bartemes KR, Kephart GM, Fox SJ, Kita H. Enhanced innate type 2 immune response in peripheral blood from patients with asthma. *J Allergy Clin Immunol*. 2014;134:671-678 e674.
102. Hams E, Armstrong ME, Barlow JL, et al. IL-25 and type 2 innate lymphoid cells induce pulmonary fibrosis. *Proc Natl Acad Sci USA*. 2014;111:367-372.
103. Spits H, Cupedo T. Innate lymphoid cells: emerging insights in development, lineage relationships, and function. *Annu Rev Immunol*. 2012;30:647-675.
104. Robinette ML, Fuchs A, Cortez VS, et al. Transcriptional programs define molecular characteristics of innate lymphoid cell classes and subsets. *Nat Immunol*. 2015;16:306-317.
105. Turner JE, Morrison PJ, Wilhelm C, et al. IL-9-mediated survival of type 2 innate lymphoid cells promotes damage control in helminth-induced lung inflammation. *J Exp Med*. 2013;210:2951-2965.
106. Huang Y, Guo L, Qiu J, et al. IL-25-responsive, lineage-negative KLRG1(hi) cells are multipotential 'inflammatory' type 2 innate lymphoid cells. *Nat Immunol*. 2015;16:161-169.
107. Gold MJ, Antignano F, Halim TY, et al. Group 2 innate lymphoid cells facilitate sensitization to local, but not systemic, TH2-inducing allergen exposures. *J Allergy Clin Immunol*. 2014;133:1142-1148.
108. Doherty TA, Soroosh P, Broide DH, Croft M. CD4+ cells are required for chronic eosinophilic lung inflammation but not airway remodeling. *Am J Physiol Lung Cell Mol Physiol*. 2009;296:L229-L235.
109. Zaiss DM, Yang L, Shah PR, Kobie JJ, Urban JF, Mosmann TR. Amphiregulin, a TH2 cytokine enhancing resistance to nematodes. *Science*. 2006;314:1746.
110. Van Dyken SJ, Mohapatra A, Nussbaum JC, et al. Chitin activates parallel immune modules that direct distinct inflammatory responses via innate lymphoid type 2 and gammadelta T cells. *Immunity*. 2014;40:414-424.
111. Paul WE, Ohara J. B-cell stimulatory factor-1/interleukin 4. *Annu Rev Immunol*. 1987;5:429-459.
112. Berton MT, Uhr JW, Vitetta ES. Synthesis of germ-line gamma 1 immunoglobulin heavy-chain transcripts in resting B cells: induction by interleukin 4 and inhibition by interferon gamma. *Proc Natl Acad Sci USA*. 1989;86:2829-2833.
113. Esser C, Radbruch A. Rapid induction of transcription of unrearranged S gamma 1 switch regions in activated murine B cells by interleukin 4. *EMBO J*. 1989;8:483-488.
114. Rothman P, Lutzker S, Cook W, Coffman R, Alt FW. Mitogen plus interleukin 4 induction of C epsilon transcripts in B lymphoid cells. *J Exp Med*. 1988;168:2385-2389.
115. Yamane H, Paul WE. Early signaling events that underlie fate decisions of naive CD4(+) T cells toward distinct T-helper cell subsets. *Immunol Rev*. 2013;252:12-23.
116. Tanaka S, Tsukada J, Suzuki W, et al. The interleukin-4 enhancer CNS-2 is regulated by Notch signals and controls initial expression in NKT cells and memory-type CD4 T cells. *Immunity*. 2006;24:689-701.
117. Phythian-Adams AT, Cook PC, Lundie RJ, et al. CD11c depletion severely disrupts Th2 induction and development in vivo. *J Exp Med*. 2010;207:2089-2096.
118. Mesnil C, Sabatel CM, Marichal T, et al. Resident CD11b(+)Ly6C(-) lung dendritic cells are responsible for allergic airway sensitization to house dust mite in mice. *PLoS ONE*. 2012;7:e53242.
119. Plantinga M, Guilliams M, Vanheerswynghels M, et al. Conventional and monocyte-derived CD11b(+) dendritic cells initiate and maintain T helper 2 cell-mediated immunity to house dust mite allergen. *Immunity*. 2013;38:322-335.
120. Williams JW, Tjota MY, Clay BS, et al. Transcription factor IRF4 drives dendritic cells to promote Th2 differentiation. *Nat Commun*. 2013;4:2990.
121. Kumamoto Y, Linehan M, Weinstein JS, Laidlaw BJ, Craft JE, Iwasaki A. CD301b(+) dermal dendritic cells drive T helper 2 cell-mediated immunity. *Immunity*. 2013;39:733-743.
122. Gao Y, Nish SA, Jiang R, et al. Control of T helper 2 responses by transcription factor IRF4-dependent dendritic cells. *Immunity*. 2013;39:722-732.
123. Bajana S, Roach K, Turner S, Paul J, Kovats S. IRF4 promotes cutaneous dendritic cell migration to lymph nodes during homeostasis and inflammation. *J Immunol*. 2012;189:3368-3377.
124. Vander Lugt B, Khan AA, Hackney JA, et al. Transcriptional programming of dendritic cells for enhanced MHC class II antigen presentation. *Nat Immunol*. 2014;15:161-167.
125. Tussiwand R, Everts B, Grajales-Reyes GE, et al. Klf4 expression in conventional dendritic cells is required for T helper 2 cell responses. *Immunity*. 2015;42:916-928.
126. Ochiai S, Roediger B, Abtin A, et al. CD326(lo)CD103(lo)CD11b(lo) dermal dendritic cells are activated by thymic stromal lymphopoietin during contact sensitization in mice. *J Immunol*. 2014;193:2504-2511.
127. Raymond M, Rubio M, Fortin G, et al. Selective control of SIRP-alpha-positive airway dendritic cell trafficking through CD47 is critical for the development of T(H)2-mediated allergic inflammation. *J Allergy Clin Immunol*. 2009;124:1333-1342 e1331.
128. Park CS, Lee PH, Yamada T, et al. Kruppel-like factor 4 (KLF4) promotes the survival of natural killer cells and maintains the number of conventional dendritic cells in the spleen. *J Leukoc Biol*. 2012;91:739-750.
129. Kitajima M, Ziegler SF. Cutting edge: identification of the thymic stromal lymphopoietin-responsive dendritic cell subset critical for initiation of type 2 contact hypersensitivity. *J Immunol*. 2013;191:4903-4907.
130. Halim TY, Steer CA, Matha L, et al. Group 2 innate lymphoid cells license dendritic cells to potentiate memory TH2 cell responses. *Nat Immunol*. 2016;17:57-64.
131. Hepworth MR, Monticelli LA, Fung TC, et al. Innate lymphoid cells regulate CD4+ T-cell responses to intestinal commensal bacteria. *Nature*. 2013;498:113-117.

132. Van Dyken SJ, Nussbaum JC, Lee J, et al. A tissue checkpoint regulates type 2 immunity. *Nat Immunol*. 2016;17:1381-1387.
133. Endo Y, Hirahara K, Iinuma T, et al. The interleukin-33-p38 kinase axis confers memory T helper 2 cell pathogenicity in the airway. *Immunity*. 2015;42:294-308.
134. Guo L, Huang Y, Chen X, Hu-Li J, Urban JF Jr, Paul WE. Innate immunological function of TH2 cells in vivo. *Nat Immunol*. 2015;16:1051-1059.
135. Jacobsen EA, Zellner KR, Colbert D, Lee NA, Lee JJ. Eosinophils regulate dendritic cells and Th2 pulmonary immune responses following allergen provocation. *J Immunol*. 2011;187:6059-6068.
136. Jacobsen EA, Doyle AD, Colbert DC, et al. Differential activation of airway eosinophils induces IL-13-mediated allergic Th2 pulmonary responses in mice. *Allergy*. 2015;70:1148-1159.

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