Cytokine 98 (2017) 59–70

Contents lists available at ScienceDirect

Cytokine

journal homepage: www.journals.elsevier.com/cytokine

Review article

Allergic diseases: From bench to clinic - Contribution of the discovery of interleukin-5

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ARTICLE INFO

Article history:
Received 28 October 2016
Accepted 18 November 2016

Keywords:
Allergy
Eosinophil
ILC2
Severe asthma
Th2
Mepolizumab/Reslizumab
Benralizumab

ABSTRACT

T helper 2 cells produce a number of cytokines including inteleukin (IL)-5, IL-4 and IL-13. Group 2 innate lymphoid cells (ILC2s) also produce IL-5 under sterile conditions. IL-5 is interdigitating homodimeric glycoprotein and a member of the four α helical bundle motifs conserved among hematopoietic cytokines. IL-5 exerts its effects on target cells via IL-5 receptor (IL-5R), composed of an IL-5R α and βc subunit. The membrane proximal proline-rich motif of the cytoplasmic domain of both IL-5R α and βc subunits is essential for IL-5 signal transduction. Although IL-5 was initially identified by its ability to support the growth and terminal differentiation of mouse B cells into antibody-secreting cells, recombinant IL-5 exerts pleiotropic activities on various target cells. For example, IL-5 is now recognized as the major maturation and differentiation factor for eosinophils in mice and humans. Overexpression of IL-5 in mouse significantly increases eosinophil numbers and antibody levels *in vivo*, while mice lacking a functional gene for IL-5 or IL-5R display developmental and functional impairments in B cell and eosinophil lineages. In mice, the role of the IL-5/IL-5R system in the production and secretion of Immunoglobulin (Ig) M and IgA in mucosal tissues has been reported. Although eosinophils protect against invading pathogens including virus, bacteria and helminthes, they are also involved in the pathogenesis of various diseases, such as food allergy, asthma, and inflammatory bowel diseases. The recent expansion in our understanding in the context of IL-5 and IL-5-producing ILC2s in eosinophil activation and the pathogenesis of eosinophil-dependent inflammatory diseases has led to advances in therapeutic options. A new therapy currently under invetigarion in clinical trials uses humanized monoclonal antibodies against IL-5 or the IL-5R. In this review, we summarize our current understanding of the functions of IL-5 and its receptor, the innate regulation of IL-5-producing cells, and therapeutic potential of anti-IL-5 and anti-eosinophil (IL-5R) antibodies.

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Abbreviations: ADCC, antibody-dependent cell-mediated cytotoxicity; ASC, antibody secreting cell; AHR, airway hyperresponsiveness; βc, common β; BCGF, B cell growth factor; BCDF, B cell differentiation factor; BCL1, murine chronic B leukemia; Btk, Bruton's tyrosine kinase; COPD, chronic obstructive pulmonary disease; DNP, 2,4-dinitrophenylated; EDF, eosinophil differentiation factor; IL, interleukin; IL-5R, IL-5 receptor; ILC2, group 2 innate lymphoid cell; Ih2, innate helper 2; Ig, imunoglobulin; JAK, janus kinase; mAb, monoclonal antibody; PAH, pulmonary arterial hypertension; STAT5, signal transducer and activator of transcription 5; Th, T helper; TRF, T cell-replacing factor; VIP, vasoactive intestinal peptide.

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http://dx.doi.org/10.1016/j.cyto.2016.11.011
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1. Introduction

Around 1970, many immunologists were interested in how T cell facilitates B cell differentiation into antibody-secreting cells (ASC). They found that T-cell-derived soluble mediators enhanced antibody response in an major histocompatibility complex-non-restricted manner [1] termed "T cell-replacing factor" (TRF). In the 1980s, numerous attempts were made to identify the molecular nature of T-cell derived cytokines including TRF that were involved in activated B cell differentiation in the absence of T cells. We previously analyzed the roles of a T-cell-derived cytokine, which we called "Enhancing factor on anti-hapten antibody response" [2] and later "TRF" on anti-hapten Immunoglobulin (Ig) G response [3]. We developed a monoclonal antibody (mAb) against mouse TRF [4] and isolated cDNA encoding the TRF active molecule [5]. As recombinant mouse TRF exerts pleiotropic activities on various target cells beside B cells, we proposed calling TRF "interleukin (IL)-5" [5,6]. Recombinant IL-5 activates mouse B cells and eosinophils and induces their proliferation and differentiation. In humans, the biological effects of IL-5 have been best characterized using eosinophils [7,8].

IL-5 acts on target cells by binding to its specific IL-5 receptor (IL-5R). In mice, IL-5 responsive B cells and eosinophils express small numbers of high-affinity IL-5R and large numbers of low-affinity IL-5R. The biological responsiveness of IL-5 depends on interactions with the high-affinity IL-5R composed of IL-5R α and common β (βc) subunits [9–14] IL-5R α is expressed on mouse B cells and eosinophils. B cells can be subdivided into B-1 B cells and conventional B (B-2 B) cells, which regulate the innate and acquired immune responses, respectively. B-1 B cells have specific characteristics, including the constitutive expression of the high-affinity IL-5R, and differentiation into IgM-secreting cells in response to IL-5 [15,16]. Naïve B-2 B cells express low numbers of IL-5R α, if any, but fully express it upon stimulation with antigen and T helper (Th) cells. They then respond to IL-5 by differentiating into ASCs. IL-5-stimulated activated B-2 cells also undergo genetic events in their IgH gene loci for IgG1-switch recombination, where the heavy chain constant region is changed from Cμ to Cγ1 [17,18]. Although several cytokines affect eosinophil functions, IL-5 is the major cytokine involved in the regulation of blood and tissue eosinophils [7,8]. Therefore, it has been postulated that IL-5 is a therapeutic target for eosinophil-associated diseases such as asthma.

Th2 cells and mast cells are major IL-5-producing cells [6,19]. Interestingly, IL-5 expression was observed in the tissues of T cell- or mast cell-deficient mice [15]. To elucidate which cells other than Th2 cells produce IL-5 under sterile conditions, we established IL-5 reporter mice that enabled us to detect IL-5-producing cells as florescent protein Venus^{+} cells [20]. Using this system, we identified IL-5-producing non-T lymphoid cells that resided in the intestine, peritoneal cavity and lungs in naïve mice. These cells share many overlapping characteristics with natural helper cells, nuocytes and innate type 2 (Ih2) cells such as surface antigens and responsiveness to cytokines. Innate cells producing Th2 cytokine including IL-5-producing innate cells are now called group 2 innate lymphoid cells (ILC2s). Numerous studies have reported that ILC2s are involved in innate immune responses in allergy and infection [21].

Allergic diseases including asthma and atopic diseases are characterized by inflammation with the pronounced infiltration of T cells and granulocytes including mast cells, eosinophils, and neutrophils. Recruitment of CD4^{+} T cells and eosinophils is a central feature of the late-phase allergic response. Pulmonary allergen exposure results in the increased output of eosinophils from hemopoietic tissues and their increased migration to the lung [22]. T-cell-derived IL-5 and eosinophils are thought to play critical roles in the induction of airway hyperreactivity and the development of lesions that underpin chronic airway wall remodeling. In addition, thymic stromal lymphopoietin (TSLP), IL-25, and IL-33 appear to be involved in the induction of allergic inflammation in asthma.

Eosinophilia is associated with a wide variety of conditions including asthma and atopic diseases. Data from animal models support a role for IL-5 in the induction of eosinophilic inflammation. For example, the pivotal role played by IL-5 in the late asthmatic response was confirmed by the capacity for neutralizing anti-IL-5 mAb to inhibit antigen- or virus-induced airway hyperresponsiveness and eosinophil infiltration in the airways of mice and guinea pigs [23,24]. Early case reports and treatment of small cohorts of patients who have eosinophilia using anti-IL-5 mAbs—either mepolizumab or reslizumab/SCH55700—showed promising results [25–27]. Results of humanized anti-IL-5 mAb treatment in patients with mild asthma confirmed the importance of IL-5 in eosinophil-mediated inflammation in humans. However, whether long-term anti-IL-5 antibody treatment can reduce the incidence of exacerbation of eosinophilic asthma remains to be determined.

As there are already outstanding reviews providing general information on IL-5 and IL-5R, this review article briefly summarize the historical background of IL-5 research and introduce recent

advances in IL-5-producing ILC2s in the immune system and inflammation. We also report the results of recent clinical application of anti-IL-5 and anti-IL-5R antibody therapy for eosinophilic asthma.

2. IL-5/IL-5R

2.1. Cloning of IL-5

The factor that induced the terminal differentiation of B cells to Ig-secreting cells was first termed TRF, then B cell growth factor (BCGF) II or B cell differentiation factor (BCDF) which later was determined to be a prototype of IL-5 [1–6]. We observed TRF activity in the supernatants of lymph node cell cultures of *Mycobacterium tuberculosis*-primed mice after stimulation with purified protein derivatives. We monitored TRF activity by its ability to induce anti-2,4-dinitrophenylated (DNP)-IgG ASCs from T-cell-depleted DNP-primed B cells in an adoptive cell transfer system and *in vitro* culture system [2,3]. To identify TRF-active molecules, we developed a TRF-producing T cell hybridoma (B151K12, B151 in short) and established a reproducible TRF assay system using *in vivo* murine chronic B-cell leukemia (BCL1) cells [28,29]. Cultured supernatants of B151 cells showed TRF activity on BCL1 cells and induced their differentiation into IgM-ASC. B151 cells do not produce detectable levels of other cytokines, but constantly produce TRF without exogenous stimulation. HPLC-purified B151-derived TRF (B151-TRF) is an acidic glycoprotein that exerts BCGF II activity causing BCL1 cells or dextran sulfate-stimulated splenic B-2 B cells to proliferate [29].

We generated two mAbs (NC17 and TB13) using HPLC-enriched B151-TRF as the immunogen [4]. NC17 and TB13 neutralized the TRF and BCGFII activities of B151-TRF, but did not neutralize recombinant IL-1, IL-2, IL-3, IL-4 or interferon-γ activities. B151-TRF can be purified using a TB13 mAb-coupled immunoaffinity column. Purified B151-TRF has a molecular mass of 50–60 kDa under non-reducing conditions. The reduced and alkylated TRF of 25–30 kDa does not show any other cytokine activity, indicating that T-cell-derived TRF consists of homodimers and that dimer formation is essential to exsert its biological activities.

To elucidate the molecular mechanisms of TRF, Kinashi et al. isolated cDNA encoding mouse TRF using an expression vector system (pSP6K) containing the SP6 promoter from cDNA libraries constructed from the poly(A)$^+$ RNA of an allo-reactive T-cell clone, 2.19 [5,30]. Recombinant mouse TRF exerts both TRF and BCGFII activities which are completely blocked by our anti-TRF mAb, demonstrating that a single molecule is responsible for both TRF and BCGFII activities. Although mouse TRF was originally thought to only active B cells, recombinant TRF increases the expression of IL-2R α on antigen-stimulated thymocytes and induces their differentiation into cytotoxic T lymphocytes in the presence of IL-2 [30]. Furthermore, TRF plays critical roles in the differentiation of hematopoietic progenitors into eosinophils and in the activation of mature eosinophils [31,32]. Because TRF has similar activity of the fifth interleukin, we proposed calling TRF "IL-5". IL-5 is a synonym for the following cytokines: TRF, BCGFII, IgA-enhancing factor, IL-2Rα-inducing factor, killer helper factor, and eosinophil differentiation factor (EDF) and eosinophil colony-stimulating factor. Using mouse IL-5 cDNA as a probe, human IL-5 cDNA was isolated from a cDNA library constructed with poly(A)$^+$ mRNA extracted from ATL-2 cells [33]. Both the mouse and human IL-5 genes consist of three introns and have numerous repetitive sequences [34]. A cDNA clone encoding human EDF was independently isolated by other groups and the entire amino acid sequence is identical to that of human IL-5.

The functional assessment of mouse and human recombinant IL-5 has convincingly demonstrated that IL-5 is a disulfide-linked homodimers with a molecular mass of 50–60 kDa [5,6]. The large variation in the molecular weight of IL-5 is predominantly caused by the heterogeneous addition of carbohydrates in posttranslation [35]. Human IL-5 shares 70% sequence homology with mouse IL-5 at the amino acid level.

Crystal structure analysis of human IL-5 revealed that IL-5 is a unique member of the short-chain helical-bundle subfamily of cytokines whose canonical motif contains four helices (A-D) arranged in an up-up-down-down topology [36]. The tertiary structure of human IL-5 might explain why it absolutely requires dimerization to mediate its biological activities.

2.2. Characteristics of IL-5R

To elucidate how IL-5 exerts multiple functions in various target cells, the molecular and biochemical characteristics of IL-5R were extensively analyzed. A series of IL-5 binding and cross-linking studies revealed that murine IL-5 binds to cell surface IL-5R, which consists of two distinct subunits, α and β, with molecular masses of 60 kDa and 130 kDa, respectively [37,38]. IL-5 specifically binds to the IL-5Rα subunit [39]. We developed two anti-IL-5Rα mAbs, H7 and T21, which recognize 60 kDa proteins on IL-5-responsive cells and specifically inhibit the binding of IL-5 to IL-5R and IL-5-dependent cell proliferation [40]. Rolink et al. generated another anti-IL-5R mAb, R52.120, which partially inhibits the IL-5-induced proliferation of BCL1 cells and recognizes 130 kDa proteins. Aic2B, anti-IL-3R mAb, shows similar inhibitory activity to that of R52.120; therefore, we speculated that H7 and R52.120 or Aic2B recognized IL-5Rα and IL-5Rβ, respectively.

Expression cloning of cDNA encoding the mouse IL-5Rα subunit on an IL-5-dependent early B cell line was performed using anti-mouse IL-5Rα mAb [9]. Nucleotide sequencing of mouse IL-5Rα cDNA revealed that mouse and human IL-5Rα is a type I transmembrane protein. The extracellular domain of the mouse IL-5Rα contains two motifs—four cysteine residues and WSXWS motifs located close to the transmembrane domain—that are conserved in a set of type I cytokine receptor family members. The cytoplasmic domain has a motif rich in proline following the transmembrane domain.cDNAs of the human IL-5Rα subunit have been also isolated from human eosinophils and a cell line, HL60 [11]. The entire nucleotide sequence of human IL-5Rα cDNAs has considerable similarity to the coding sequence of mouse IL-5Rα and retains characteristics common to the type I cytokine receptor superfamily. The cytoplasmic regions rich in proline residues following the transmembrane domain are well conserved. It still remains unclear whether IL-5Rα is expressed on human B cells localized in various tissues and whether IL-5 is required for the activation and antibody production by human intestinal B cells. IL-5Rα is expressed on eosinophils and basophils in humans and mice.

COS7 transfectants expressing mouse IL-5Rα bind IL-5 with low-affinity but they do not respond to IL-5. However, the enforced expression of mouse IL-5Rα in IL-3-dependent FDC-P1 cells enabled them to reconstitute high affinity IL-5Rα and induce IL-5-dependnent proliferation, indicating that IL-3-dependent FDC-P1 cells constitutively express the mouse IL-5Rβ subunit. Transfection of *Aic2B* cDNAs into IL-5Rα-expressing L cells induced high-affinity IL-5Rα expression. Furthermore, enforced expression of mouse IL-5Rα and AIC2B in CTLL enabled them to proliferate in response to IL-5. Because AIC2B does not bind IL-5 and forms high-affinity IL-5R in combination with IL-5Rα [10], we concluded that AIC2B is the mouse IL-5Rβ subunit and has an indispensable role in IL-5 signal transduction. In humans, KH97, a homologue of mouse AIC2 could reconstitute high-affinity human IL-5R with human IL-5Rα and transduce IL-5 signals. KH97 is the human IL-5Rβ, known as βc, has been reported to be the β subunit of human IL-3R and human granulocyte-macrophage colony-

stimulating factor receptor [10–12,39]. The membrane proximal proline-rich motif of IL-5Rα and βc is indispensable for IL-5 signal transduction [9,10,41–43].

2.3. IL-5R signaling

Crystal structure analysis has clearly demonstrated that the IL-5 dimer binds to IL-5Rα in a wrench-like structure with an octamer-motif of IL-5/IL-5Rαβ complexes [44,45]. This binding is required for signal transduction through IL-5Rα and induces rapid tyrosine phosphorylation of various signaling molecules such as Janus kinase 1 (JAK1), JAK2, signal transducer and activator of transcription 5 (STAT5), phosphoinositide 3-kinase and mitogen-activated protein kinases [18,41,42,46–48]. In human eosinophil as well as mouse eosinophil and B cells, the activation of JAK2 and STAT5 is indispensable for IL-5-dependent signal transduction [18,42,43,46–49]. In addition to these molecules, the Ras GTPase-extracellular signal-regulate kinase pathway contributes to IL-5-dependent cell survival, proliferation and differentiation of eosinophils [50,51].

We previously reported that tyrosine phosphorylation and activation of Bruton's tyrosine kinase (Btk) is required for an IL-5-dependent mouse B cell activation. Btk is a cytoplasmic tyrosine kinase expressed in myeloid, erythroid, and B-lineage cells except plasma cells whose activation is essential for B-cell development in both mice and humans. B cells from *Btk*$^{-/-}$ mice have impaired B cell development and are hyporesponsive to IL-5 and IL-10. However, Btk appears not to be essential for IL-5-dependent eosinophil maturation and activation (our unpublished data).

2.4. Functions of IL-5 in B cells

IL-5 was originally identified as a factor that induced the differentiation of BCL1 cells into IgM-secreting cells and DNP-primed B cells into anti-DNP IgG producing cells. Thus, the functions of IL-5 in mouse B cells are well characterized. IL-5 can induce both antigen-specific IgA production from antigen-primed B-2 B cells and polyclonal IgA from lipopolysaccharide-stimulated B-2 B cells [52–57]. Further analyses revealed that IL-5 affects surface (s) IgA^{+} B-1 B and B-2 B cells but not sIgA^{-} B-2 B cells for the induction of IgA-producing cells [55,56]. IL-5 does not induce the expression of germ-line Cα transcripts or the formation of IgA-specific switch circular DNA, indicating it is not a class-switching factor for μ to α. Rather, IL-5 induces the terminal differentiation of sIgA^{+} B-2 B cells into IgA-producing cells. Transforming growth factor-β is a class-switching factor for μ to α [56]. In addition, IL-5 induces the expression of IL-2Rα on B-2 B cell [58–60].

As described earlier, IL-5Rα is constitutively expressed on murine B-1 B cells but is expressed at low levels on murine B-2 B cells in the spleen [40,61]. Using IL-5Rα-deficient mice, we demonstrated that IL-5 signaling was indispensable for murine B-1 B cell proliferation, survival, and mucosal IgA production [57,62]. Therefore, IL-5 has pivotal roles in the homeostasis and immune responses of murine B-2 B and B-1 B cells [63].

2.5. Functions of IL-5 in eosinophils

Eosinophils are closely related to health and disease [22]. Eosinophils have important roles in protective immune responses against invading pathogen, such as virus, bacteria and especially helminthes, by producing major basic protein, eosinophil cationic proteins, and eosinophil peroxidase. Previous studies proposed that eosinophils might be involved in tumor surveillance via their influx into tumor-growth sites [20,64,65]. It was reported that eosinophils maintain plasma cells in murine bone marrow and the intestine [66,67], indicating eosinophils might contribute to homeostasis via various mechanisms.

There is a considerable body of evidence demonstrating that eosinophils have a significant impact on allergy and asthma. Massive eosinophilia in tissues, blood, and bone marrow is one of the major features of asthma. The involvement of eosinophils in asthma was clearly demonstrated using two kinds of eosinophil-deficient mice (PHIL and Δdbl-GATA) [68–70]. Animal models support a role for IL-5 in eosinophil-associated diseases. In IL-5-transgenic mice, the massive expansion of eosinophils in the peripheral blood and bone marrow was observed [71,72]. A role for IL-5 in late asthmatic responses was demonstrated using the anti-IL-5 antibody, NC17. Other studies showed that anti-IL-5 mAbs inhibited antigen-induced airway hyperresponsiveness and eosinophil influx in the airways [24,73–75]. In respiratory antigen-challenged IL-5-deficient or IL-5Rα-deficient mice, bronchial hyperreactivity and eosinophila were not observed in the lungs compared with wild-type mice [23,76,77]. Therefore, IL-5 might be a therapeutic target for eosinophil-mediated inflammation.

3. ILC2s and innate IL-5-producing ILC2s

3.1. Identification and characterization of ILC2s

Beyond classical Th2 cells, the presence of previously unknown IL-5 producing cells has been suggested. Additionally, it has been reported that type 2 immune responses can be triggered in the absence of T cells [78,79]. In 2010, a non-B/non-T cell population was defined by independent laboratories as either nuocytes [80], Ih2 cells [81], or natural helper cells [82]. These types of cells are now collectively referred to as ILC2s [83]. Flow cytometry analysis revealed that ILC2s were enriched in lineage marker negative cells and express c-kit, Sca-1, Thy1, and ICOS. Importantly, ILC2s express cytokine receptors including CD127 (IL-7Rα), CD25 (IL-2Rα), IL-17RB (an IL-25 receptor subunit), ST2 (an IL-33 sreceptor subunit), and TSLPR [83]. Because IL-25 and IL-33 have critical roles in the production of Th2 cytokines [84], ILC2s might be a potent cellular source of Th2 cytokines.

3.2. IL-5-producing ILC2s

We independently investigated innate IL-5-producing cells in resident tissues under physiological settings. To this end, we generated an IL-5/Venus knockin mouse in which IL-5-producing cells are easily detected by flow cytometry and immunohistochemistry [20]. IL-5-producing Venus^{+} cells were present in the lamina propria of the small and large intestines, peritoneal cavity, and the lung, but not other immunological tissues including the bone marrow, spleen, lymph nodes, mesenteric lymph nodes, and Peyer's patches from naïve mice. As for ILC2s, the Venus^{+} cells were enriched in lineage negative cells expressing c-Kit, Sca-1, Thy1, CD25, CD127, and ST2. This suggests that the majority of Venus^{+} cells overlap with the population of ILC2s and marks IL-5-producing ILC2s in the steady state [20]. Furthermore, two-thirds of Venus^{+} cells in the lung expressed intracellular IL-5 but not IL-13. In addition, IL-4-producing cells were rarely detected in Venus^{+} cells. In agreement with this finding, ILC2s were reported to produce low levels of IL-4 and in a mouse asthma model, most IL-4 was secreted from Th2 cells [85,86]. From these results, we named the Venus^{+} cells detected in our IL-5-reporter mice as innate IL-5-producing ILC2s.

3.3. Function and regulation of IL-5-producing ILC2s

ILC2s differentiate from a common ILC progenitor in the bone marrow [87,88]. The development of ILCs2 is tightly regulated by

transcriptional factors, including RORα [89,90], GFI-1 [91], GATA3 [88,92], TCF1 [93,94], and Bcl11b [94,95]. Because innate IL-5-producing ILC2s express receptors for IL-2, IL-7, IL25, and IL-33, we stimulated pulmonary cells with these cytokines and investigated the proliferative response and IL-5 production *in vitro* [20]. IL-33 stimulation potently induced the proliferation of IL-5-producing ILC2s and the production of relatively high levels of IL-5 compared with other cytokines [20,82,88]. IL-25 stimulation exhibited similar effects, but to a lesser extent compared with IL-33 stimulation [20]. *In vivo* analyses also revealed that the passive administration of IL-25 and IL-33 into mice induced the proliferation of IL-5-producing ILC2s in the lung, leading to the infiltration of eosinophils in the lung [20]. Of interest, IL-33 dramatically increased the number of IL-5-producing ILC2s in the lung. Thus, IL-33 has critical roles in the regulation of IL-5-producing ILCs2 and the subsequent accumulation of eosinophils in the lung. In addition, IL-9 has been linked to Th2 immune responses [96]. ILC2s express IL-9R [79,97] and are a major source of IL-9 [97–100], which regulates cell survival and cytokine production by ILC2s in an autocrine fashion. In the literature, IL-9 production by ILC2s promoted the rapid production of IL-5 and IL-13 production and had a critical role in tissue repair in the lung [100].

It is important to clarify how ILC2s sense tissue damage, infection, and the disruption of tissue homeostasis, even though ILC2s do not directly recognize pathogen-associated molecular patterns via innate immune sensors such as TLRs. Soluble mediators such as cytokines, metabolites, and peptides may activate ILC2s. All-trans retinoic acid, a functional metabolite of vitamin A, binds heterodimeric RAR or RXR receptors, which are expressed by ILC2s [101]. In mice, retinoic acid suppresses ILC2 responses, and vitamin A-deficient mice displayed a potent anti-helminth response [102]. Intriguingly, ILC2s may functionally interact with the nervous system. Vasoactive intestinal peptide (VIP), a neuropeptide, is secreted by enteric neurons and activates ILC2s. The expression of VIP is regulated by circadian rhythm [103], indicating that ILC2 activation and eosinophil accumulation in local tissues are controlled by the circadian clock.

The long-lasting activation of ILC2s may disrupt tissue homeostasis, but how ILC2 responses are negatively regulated remains unknown. Some mediators including cytokines may inhibit ILC2 responses by promoting a different type of immune reaction or the resolution of inflammation. For example, type 1 and 2 interferons inhibit ILC2 activation [104–106], which consequently promote type 1 and type 2 immunity. ILC2 responses are also inhibited by the soluble mediators maresin and lipoxin A4, which resolve inflammation. Maresin induces the generation of Treg cells, which limit ILC2 responses [107] and lipoxin A4 signals through its receptor on ILC2s and decreases IL-13 production [108].

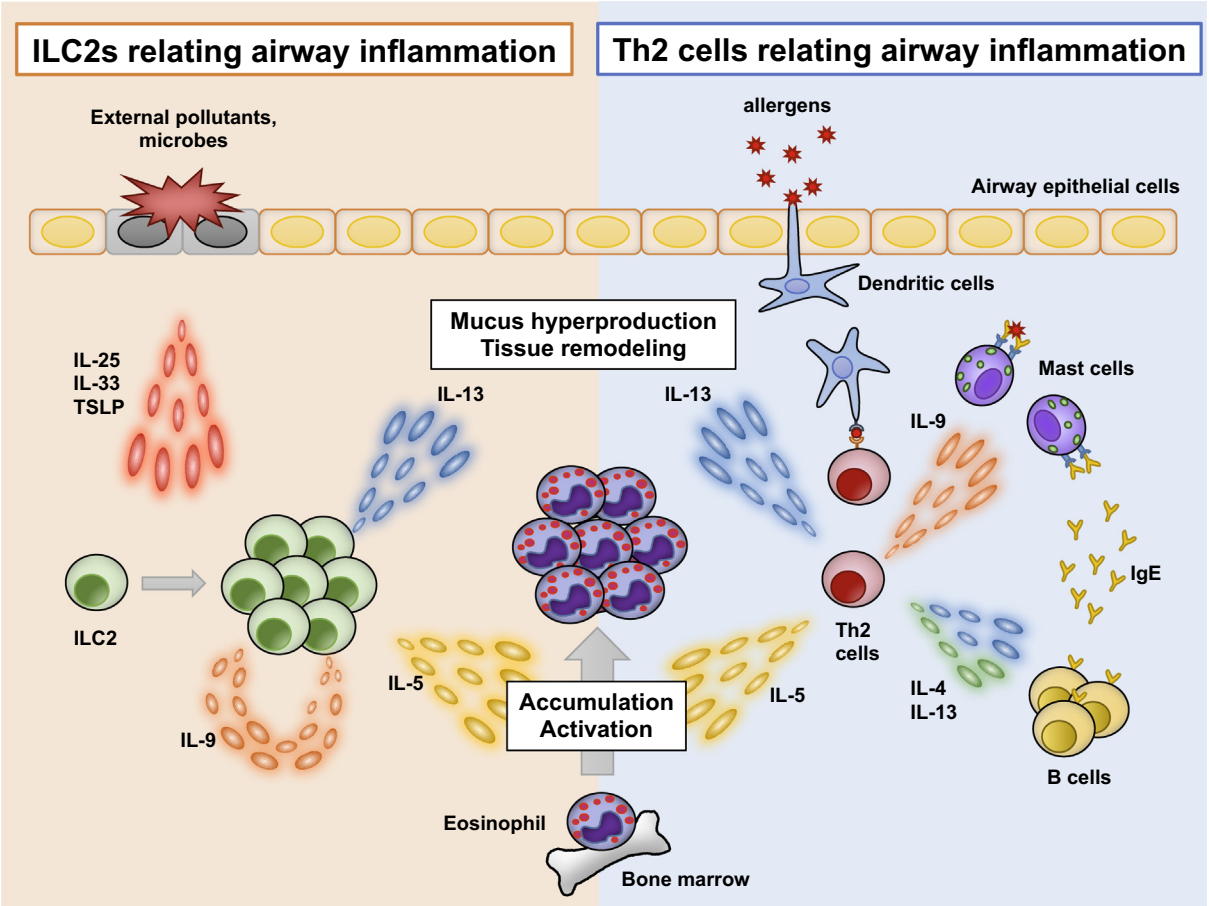


Fig. 1. The roles of Th2 cells and ILC2s in airway inflammation. Th2 cells play a key role in allergen-specific respiratory inflammation (right). Dendritic cells process antigens and present them to T cells. These T cells differentiate into Th2 cells and produce IL-4 and IL-13, leading to IgE production by B cells. These IgE bound to mast cells. When IgE binding mast cells are exposed to same antigens, IgE on mast cells and antigens form a cross-linking and degranulation of mast cells are induced. IL-9 released by Th2 cells induces mast cell expansion. On the other hand, environmental stimuli, including pollutants and infections of bacteria, virus and helminthes trigger ILC2s relating airway inflammation (left). These stimuli induce airway epithelial cell death and productions of IL-25, IL-33 and TSLP. These cytokines promote massive expansion of ILC2s. ILC2s produce Th2 cytokines, including IL-5, IL-9 and IL-13. IL-9 produced by ILC2s mediates survival of ILC2s in an autocrine manner. IL-5 and IL-13 released by both Th2 cells and ILC2s are involved in the activation/proliferation of eosinophils and mucus hypersecretion/tissue remodeling, respectively.

3.4. ILC2s and IL-5-producing ILC2s in IgA production

Several *in vitro* studies indicate that IL-5 enhances IgA production by activated B cells either alone or in combination with transforming growth factor β [54–56]. In addition, passive administration of IL-5 into wild-type mice or constitutive production of IL-5 in IL-5-transgenic mice has been shown to enhance IgA production [71]. However, serum and fecal IgA levels in *Il5rα*$^{-/-}$ mice are comparable to those of wild-type mice [43,62]. This discrepancy has not been clearly elucidated. There are two pathways of IgA production, T cell-dependent (TD) and T cell-independent (TI) pathways [109]. Using IL-5/Venus knockin mice, we demonstrated that the proportion of IL-5-producing ILC2s was small in Peyer's patches, where TD IgA takes place [20]. Therefore, it is likely that IL-5-producing ILC2s contribute to TI IgA production. Our unpublished data suggest that TI IgA production is dependent at least in part on IL-5-producing ILC2s. The frequency of TI IgA-producing cells in the intestine was only ten percent of total IgA-producing cells. This may account for the fact that we do not observe significant differences in serum and fecal IgA levels between wild-type and *Il5rα*$^{-/-}$ mice. Chu et al. demonstrated that eosinophils contributed IgA production in murine intestine through the maintenance of plasma cells [67]. In addition, eosinophils promote B cell survival, proliferation, and Ig production in mice and humans [110]. Therefore, an IL-5-producnig ILC2-eosinophil axis might contribute to TI IgA production in the intestine.

3.5. ILC2s and IL-5-producing ILC2s in allergic asthma

Th2 cytokines including IL-4, IL-5, and IL-13 are all tightly linked to the pathogenesis of allergic inflammation [111] (Fig. 1). IL-4 induces IgE production, which binds to its high affinity receptor FcεR1 on mast cells and basophils [7,112]. IL-5 promotes eosinophil accumulation in local tissues where they secrete various inflammatory mediators [7]. IL-13 affects the airway epithelium and smooth muscle cells and contributes to mucus hyperproduction and tissue remodeling [113]. Data obtained from animal models have suggested that ILC2s have a detrimental link to allergic diseases such as asthma, atopic dermatitis, and chronic rhinosinusitis. Furthermore, levels of Th2 cytokines derived from ILC2s are often much higher than that derived from Th2 cells [114]. Therefore, ILC2s and IL-5-producing ILC2s might be attractive therapeutic targets for the treatment of these diseases. Here, we will describe our recent ideas regarding the pathological roles of ILC2s in allergic inflammation, in particular allergic asthma.

Allergic asthma is a chronic airway inflammatory disease characterized by airway hyperreactivity (AHR), bronchoconstriction, increased mucus secretion and limited airflow. Protease allergens, including papain and house dust mite, are used to trigger asthma-like airway inflammation. ILC2-deficient *Rag2*$^{-/-}$/*Il2rg*$^{-/-}$ mice restored papain-induced asthmatic symptoms such as eosinophilia compared with ILC2-sufficient *Rag2*$^{-/-}$ mice [115]. Reconstitution of ILC2s in *Rag2*$^{-/-}$/*Il2rg*$^{-/-}$ mice induced asthmatic symptoms in a papain-mediated asthma model. ILC2s triggered AHR in non-infectious airway inflammation and infections such as influenza virus and fungus. The fungal allergen *Alternaria alternata* activated ILC2s and increased the production of IL-5, IL-13, and IL-9 [80,98,116]. IL-13 is sufficient to induce AHR in cytokine-based models of pulmonary inflammation. Furthermore, *Il13*$^{-/-}$ mice had reduced AHR after IL-33 or IL-25 inhalation compared with wild-type mice, which was rescued by the adoptive transfer of wild-type ILC2s [85].

Recent reports suggested that ILC2s functionally interact with other innate immune cells in allergic diseases. M2 macrophages are considered anti-inflammatory cells but have a role in pathogenic Th2 response [117]. Van Dyken et al. reported that ILC2-derived IL-13 contributed to M2 macrophage accumulation in the lung in chitin-induced allergic inflammation. In the human lung and mouse skin, ILC2s closely localize with mast cells [118]. These ILC2s constitutively express IL-13, and ILC2-derived IL-13 suppresses tumor necrosis factor and IL-6 production by mast cells, suggesting that ILC2s negatively regulate mast cell function via IL-13. One study reported that ILC2s interact with basophils in inflamed human and mouse skin [119] and clusters of ILC2s and basophils were observed in these skin lesions. Furthermore, basophil-derived IL-4 plays a role in the recruitment of ILC2s to atopic dermatitis skin lesion. However, it remains unclear whether basophils regulate ILC2 functions in allergic inflammation in the lung.

Recent studies showed that ILC2s are associated with asthma, in particular severe asthma. In steroid resistant asthma, high levels of ILC2s were observed in sputum compared with healthy individuals and mild asthmatic patients [120]. Furthermore, the number of ILC2s in the sputum of severe asthma patients was increased compared with pulmonary infection patients without asthma [121]. Notably, the numbers of ILC2s in peripheral blood mononuclear cells were significantly high in allergic patients compared with healthy individuals and these cells produced large amounts of IL-5 and IL-33 in response to IL-25 or IL-33 stimulation [122]. Additionally, several genome-wide studies have suggested that genetic variants of IL-33, ST2, TSLP, and IL-17RB are associated with an increased susceptibility to asthma [123–126]. Therefore, ILC2s might be a key player in the pathogenesis of severe asthma and be a potential therapeutic target for this disease.

3.6. ILC2s and IL-5-producing ILC2s in other lung diseases

Chronic obstructive pulmonary disease (COPD) is characterized by progressive bronchitis, emphysema and limited airflow in the lungs. Various inhaled particles and gases, such as cigarette smoke, trigger the development of COPD, and viral infection can exacerbate COPD [127]. Intriguingly, respiratory viral infection augments eosinophilia in the bronchoalveolar lavage and increases the expression of IL-5 and IL-13 mRNA in the lung [128]. Because viral infection induces IL-33 release from epithelial cells, IL-33-activated ILC2s may exacerbate the symptoms of COPD by enhancing IL-5 and IL-13 expression and eosinophilia in the lung.

It has been suggested that eosinophils promote tumorigenesis or anti-tumorigenesis [129–131]. Deficiency of the IL-5 signal causes impaired eosinophil recruitment in the lung; therefore, we investigated the roles of eosinophils in lung melanoma metastasis using IL-5-deficient mice (IL-5$^{V/V}$ mice) and IL-5Rα-deficient mice [20]. Increased tumor metastasis was observed in IL-5- or IL-5Rα-deficient mice compared with wild-type mice. Recombinant IL-5 injection induced eosinophil influx in the lung and prolonged migrated eosinophil metastatic melanoma cells [20]. In addition, melanoma injection promoted the transient upregulation of IL-33 mRNA and prolonged the expression of IL-5 mRNA in the lung. Taken together, IL-5-producing ILC2s and IL-5 in the lung may have a protective role against tumor metastasis via the ILC2-eosinophil axis.

Pulmonary arterial hypertension (PAH) is characterized by severe obstruction of small pulmonary artery, leading to the development of high pulmonary artery pressure. A previous paper reported that levels of soluble ST2 in sera were increased in PAH patients [132], suggesting that IL-33 might be associated with the pathogenesis of PAH. Furthermore, IL-5 is involved in the development of arterial hypertrophy in an animal model of PAH [133]. Since PAH was triggered by Th2 responses in ovalbumin- or house dust mite extract-induced model [134–136], Th2 cells were suggested to be major IL-5 producers in these PAH models. In contrast, our

unpublished data revealed that IL-33 treatment induced severe arterial hypertrophy in the lung and the incidence of the hypertrophy was largely dependent on IL-5 production from ILC2 s. From these results, the IL-33-IL-5-producing ILC2 axis could be one of therapeutic targets of PAH.

4. Anti-IL-5 therapeutics for human diseases

4.1. Eosinophils and asthma

As already described in this article, accumulated scientific knowledge since the discovery of IL-5 have determined that the most important biological role of IL-5 is as a growth and survival factor for eosinophils in humans. Eosinophils have an important role in the development of certain allergic diseases including asthma [22,137]. In particular, increased eosinophil levels have been linked to inflammation in the airways and the exacerbation of asthma [138]. Emerging concepts in the role of IL-5 in asthma have been discussed through the integration of results from animal studies and analyses of clinical disorders.

Eosinophils are a white blood cell whose natural role is to defend the body against parasites. Eosinophils also accumulate during allergic reactions, including some types of asthma and release chemicals such as eosinophil cationic protein, eosinophil-derived neurotoxin, and eosinophil peroxidase that are very efficient at fighting parasites, but can damage the lining of the lung in patients with asthma [22,137,139]. Patients with asthma marked by eosinophilic inflammation account for about 20% of those with refractory asthma [140]. They have severe exacerbations that can only be treated with systemic corticosteroids. Despite these available therapies, some symptoms remain poorly controlled even for those receiving maximum therapy (including oral corticosteroids). Some patients with severe persistent asthma who are resistant to oral corticosteroids are at a greater risk for more frequent asthma exacerbations and hospitalizations [141,142]. They tend to have airway remodeling associated with persistent airflow limitation [143–145]. Recent large epidemiological studies suggested that the elevated blood eosinophil levels are an independent risk factor for future asthma exacerbations [138]. Therefore, the development of therapies to control asthma in this subpopulation is an important therapeutic step for improving asthma outcomes.

Gleich et al. proposed a working hypothesis almost four decades ago that eoshinophils are one of the major effector cells in the pathophysiology of asthmaby damaging respiratory epithelium through the release of cytotoxic granule proteins [146]. However, this was difficult to prove clinically until the recent development of potent IL-5 inhibitors.

4.2. Development of IL-5 antagonists

Based on accumulated scientific knowledge regarding the association between eosinophils and allergic diseases, there has been significant industrial interest in the discovery of IL-5 antagonists. Although small molecules [147], peptides [148] and proteins [149,150] with antagonistic activity against IL-5 derived from random and rational approaches were reported, mAbs against IL-5/IL-5R have been clinically successful anti-IL-5 therapeutics.

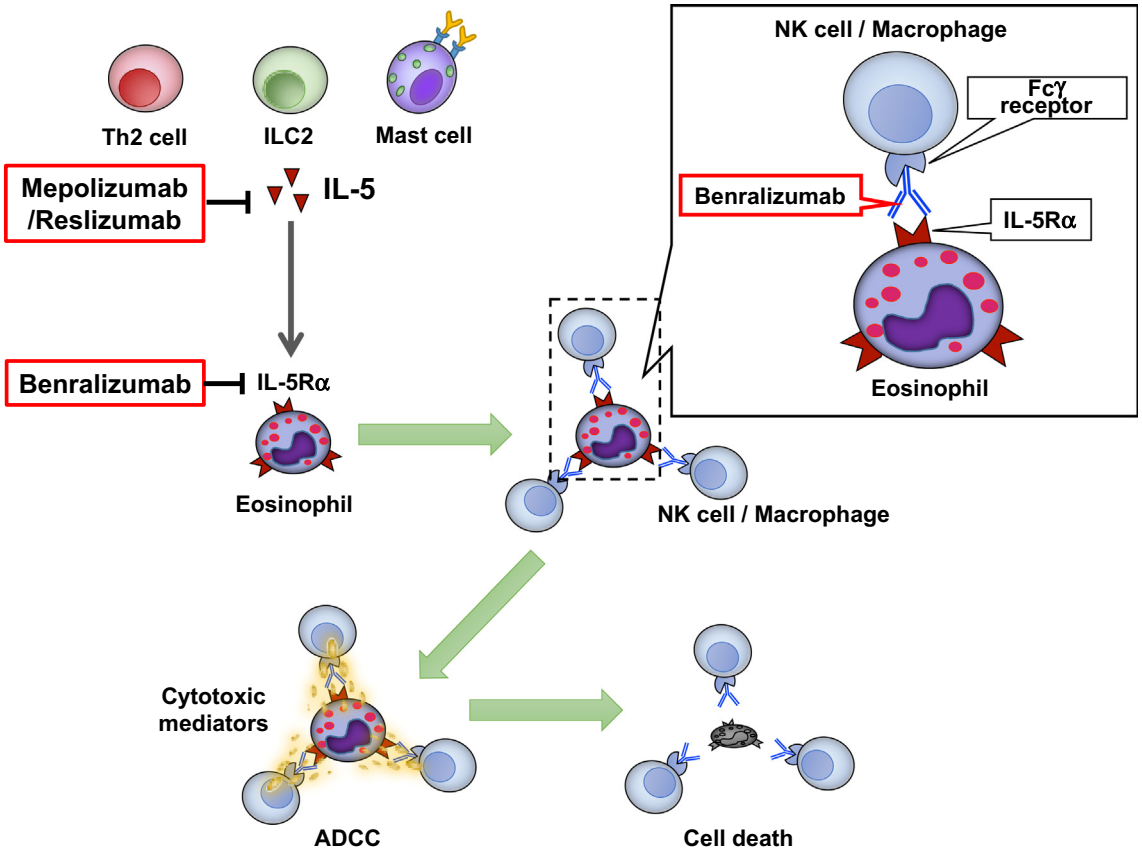


Fig. 2. Schematic illustration of ADCC against eosinophils using anti-IL-5Rα antibody. IL-5 derived from Th2 cells, mast cells and ILC2s, induces activation and proliferation of eosinophils. Mepolizumab and reslizumab neutralize IL-5 production and inhibit activation and proliferation of eosinophils. On the other hand, benralizumab acts on IL-5Rα on eosinophils and inhibits IL-5 signal transduction. In addition, benralizumab induces cell death of eosinophils by ADCC. The Fc region of benralizumab also binds to Fcγ receptor on natural killer cells and macrophages, and induces secretion of cytotoxic mediators, such as perforin and granzyme. These mediators induce eosinophil death.

Mepolizumab was the first therapeutic mAb (humanized IgG1, Kappa, mAb) to human IL-5 to be approved for any indication globally. Mepolizumab acts by preventing IL-5 from binding to IL-5Rα complex expressed on the eosinophil cell surface. This action inhibits IL-5 signaling and the over-expressing functions of peripheral blood and tissue eosinophils [25,26,151,152]. Reslizumab, another IL-5 neutralizing mAb (humanized IgG4, Kappa, mAb), was also recently approved as an IL-5 inhibiting agent for the treatment of severe asthma [27,153,154].

The first clinical studies performed with 2 humanized anti-IL-5 antagonistic antibodies, mepolizumab and reslizumab, successfully reduced the numbers of circulating and tissue-resident eosinophils, demonstrating for the first time the importance of IL-5 for eosinophil expansion and survival in humans, although the desired improvements in asthma symptoms and lung function were not observed [27,151,152]. It was only later that the first clinical data convincingly demonstrated a causal role of the IL-5/eosinophil axis in asthma exacerbations. Importantly, in these studies mepolizumab was administered to patients with severe asthma with elevated sputum eosinophil counts, who had uncontrolled disease despite the use of corticosteroids, where reduced acute exacerbations were defined as the primary clinical endpoint [25,26,155]. Multiple administrations of mepolizumab resulted in a ~50% reduction in the frequency of asthma exacerbations, improved Quality of Life Questionnaire scores and reduced the need for prednisone. These studies highlighted the benefit of selecting an appropriate clinical endpoint (acute exacerbations) to be tested in patients who presented with uncontrolled disease and elevated sputum eosinophil counts. Interestingly, although mepolizumab significantly reduced circulating and sputum eosinophil counts, its effect on reducing mucosal eosinophilia was only partial at best and did not reach statistical significance, even after prolonged high-dose exposure (12 months, 750 mg/month) [156].

4.3. Benralizumab: an anti- eosinophil monoclonal antibody with enhanced ADCC activity

Benralizumab, a mAb (glycoengineered non-fucosylated humanized IgG1, Kappa, mAb) that binds to human IL-5Rα is expected to be the first therapeutic mAb [155,157,158] to be approved as it was reported to be well tolerated and achieved the primary end-point in two pivotal Phase 3 registrational trials, demonstrating significant reductions in the annualized asthma exacerbation rate compared with placebo [159,160]. In contrast to IL-5 antagonistic mAbs, such as mepolizumab and reslizumab, benralizumab binds to cell surface IL-5Rα and induces eosinophil death through enhanced antibody-dependent cell-mediated cytotoxicity (ADCC) activity (Fig. 2). Importantly ADCC-dependent cell death of eosinophils by benralizumab treatment up-regulated the apoptosis marker annexin V on the cell surface but did not cause eosinophil degranulation *in vitro* [155], or in patients with asthma [158,161]. The distinct pharmacological activity of benralizumab is exemplified by the fast onset of eosinophil depletion in patients with asthma (within 6–24 h after administration), a profound reduction of circulating eosinophils below the limit of detection [158], the almost complete removal of lung tissue eosinophils [162], a profound effect on bone marrow eosinophils and their precursors [162] and the reduction of circulating basophils, that express IL-5Rα but do not depend on IL-5 for survival [162]. More complete depletion of eosinophils and basophils from asthmatic patients might be achieved by the treatment of benralizumab through enhanced ADCC activity. The pharmacological activity and pharmacodynamics of benralizumab make it a unique mAb for the treatment of diseases with eosinophilic inflammation such as asthma. It would be reasonable to say benralizumab might be more effective in eosinophil depletion than the mAb against IL-5. Eosinophilic inflammation has a clear role in asthma exacerbations and its depletion by benralizumab improves lung function, symptoms, and health status related to the intensity of eosinophilic inflammation [163,164].

4.4. Future development of therapeutics targeting IL-5 or IL-5Rα

After many years of research, the pivotal clinical trials of mepolizumab and reslizumab have clearly shown the benefits of IL-5 antogonists in a severe persistent asthma population uncontrolled by already-used drugs to achieve a reduction in asthma exacerbations, oral corticosteroid sparing, and a positive trend towards improvement in asthma symptoms. The two novel therapeutic antibodies targeting IL-5, mepolizumab and reslizumab, were approved on November 4th, 2015, and on March 23rd, 2016, respectively.

The first therapeutic antibody targeting IL-5Rα, Benralizumab, is expected to be approved in the near future. The hope for truly targeted therapies for uncontrolled asthma is now realized. This means that anti-eosinophil therapy may be beneficial in other eosinophil-associated diseases. Currently, clinical trials of benralizumab have been aggressively investigated for the treatment of patients suffering from COPD in addition to sever asthma. In the coming 5 years, the mechanistic role of eosinophils in the symptoms of asthma and COPD, the proportion of patients in whom the eosinophils are critical, and the magnitude of clinical efficacy of anti-eosinophil therapy will become clear. Following the development of effective therapy for asthma and COPD, attention will shift to other diseases that are dependent of eosinophilic inflammation and where eosinophils have a key role. The process to understand the relative merits of each therapy in a variety of diseases requires us to understand the physiological role of eosinophils in humans. Thus, the future development of IL-5Rα targeting therapeutics will greatly contribute to applied medicine for patients as well as to the understand of human physiology.

5. Concluding remarks

There is overwhelming evidence to support a major role for Th2 cells and ILC2s and their products in causing allergic inflammation. Among the Th2 cytokines, IL-5 has potent effects on eosinophils and induces cell maturation, survival, and activation. The two IL-5 antagonists have confirmed their clinical benefit in asthmatic patients with eosinophilic phenotype. Furthermore, anti-eosinophil mAb targeting human IL-5Rα with enhanced ADCC is considered to be a more potent eosinophil depleting reagent for the inflammatory tissues. Thus, human IL-5Rα is expected to be a successful target in development of therapeutics for eosinophilic inflammatory diseases such as asthma.

In summary, the structural, functional, and clinical studies described herein provide insight into the role of IL-5 in immune responses and the control of inflammatory diseases, and there is a strong impetus for investigating the means of modulating IL-5 effects. In concert with efficacy of anti-hIL-5 and anti-hIL-5Rα antibody therapy in hypereosinophilic syndrome and asthma, entirely new approaches and concepts for patient care should emerge in the near future.

Conflict of interest

Our laboratory (Department of Immunobiology and Pharmacological Genetics, University of Toyama) has Grant/Research support from Kyowa Hakko Kirin Co., Ltd.

Acknowledgments

We thank all members of our laboratories in the University of Toyama and Toyama Prefectural Institute for Pharmaceutical Research for helpful discussions. The authors sincerely thank Toyama Prefecture for supporting our laboratory.

This work was supported by JSPS KAKENHI Grant Numbers JP24390119 to K.T., JP15K08527 to Y.N. and JP15K18706 to T.Y., JST, PRESTO (Y.N.), Hokuriku Innovation Cluster for Health Science, MEXT Regional Innovation Cluster Program, Toyama/Ishikawa Region (K.T.) and Hokuriku Life Science Cluster, MEXT Regional Innovation Strategy Support Program (K.T.).

References

[1] A. Schimpl, E. Wecker, Replacement of T-cell function by a T-cell product, Nat. New Biol. 237 (70) (1972) 15–17.
[2] K. Takatsu, S. Haba, T. Aoki, M. Kitagawa, Enhancing factor on anti-hapten antibody response released from PPDs-stimulated tubercle bacilli-sensitized cells, Immunochemistry 11 (2) (1974) 107–109.
[3] K. Takatsu, A. Tominaga, T. Hamaoka, Antigen-induced T cell-replacing factor (TRF). I. Functional characterization of a TRF-producing helper T cell subset and genetic studies on TRF production, J. Immunol. 124 (5) (1980) 2414–2422.
[4] N. Harada, T. Takahashi, M. Matsumoto, T. Kinashi, J. Ohara, Y. Kikuchi, N. Koyama, E. Severinson, Y. Yaoita, T. Honjo, et al., Production of a monoclonal antibody useful in the molecular characterization of murine T-cell-replacing factor/B-cell growth factor II, Proc. Natl. Acad. Sci. USA 84 (13) (1987) 4581–4585.
[5] T. Kinashi, N. Harada, E. Severinson, T. Tanabe, P. Sideras, M. Konishi, C. Azuma, A. Tominaga, S. Bergstedt-Lindqvist, M. Takahashi, et al., Cloning of complementary DNA encoding T-cell replacing factor and identity with B-cell growth factor II, Nature 324 (6092) (1986) 70–73.
[6] K. Takatsu, A. Tominaga, N. Harada, S. Mita, M. Matsumoto, T. Takahashi, Y. Kikuchi, N. Yamaguchi, T cell-replacing factor (TRF)/interleukin 5 (IL-5): molecular and functional properties, Immunol. Rev. 102 (1988) 107–135.
[7] K. Takatsu, H. Nakajima, IL-5 and eosinophilia, Curr. Opin. Immunol. 20 (3) (2008) 288–294.
[8] C.J. Sanderson, Interleukin-5, eosinophils, and disease, Blood 79 (12) (1992) 3101–3109.
[9] S. Takaki, A. Tominaga, Y. Hitoshi, S. Mita, E. Sonoda, N. Yamaguchi, K. Takatsu, Molecular cloning and expression of the murine interleukin-5 receptor, EMBO J. 9 (13) (1990) 4367–4374.
[10] S. Takaki, S. Mita, T. Kitamura, S. Yonehara, N. Yamaguchi, A. Tominaga, A. Miyajima, K. Takatsu, Identification of the second subunit of the murine interleukin-5 receptor: interleukin-3 receptor-like protein, AIC2B is a component of the high affinity interleukin-5 receptor, EMBO J. 10 (10) (1991) 2833–2838.
[11] Y. Murata, S. Takaki, M. Migita, Y. Kikuchi, A. Tominaga, K. Takatsu, Molecular cloning and expression of the human interleukin 5 receptor, J. Exp. Med. 175 (2) (1992) 341–351.
[12] J. Tavernier, R. Devos, S. Cornelis, T. Tuypens, J. Van der Heyden, W. Fiers, G. Plaetinck, A human high affinity interleukin-5 receptor (IL5R) is composed of an IL5-specific alpha chain and a beta chain shared with the receptor for GM-CSF, Cell 66 (6) (1991) 1175–1184.
[13] S. Watanabe, A.L. Mui, A. Muto, J.X. Chen, K. Hayashida, T. Yokota, A. Miyajima, K. Arai, Reconstituted human granulocyte-macrophage colony-stimulating factor receptor transduces growth-promoting signals in mouse NIH 3T3 cells: comparison with signalling in BA/F3 pro-B cells, Mol. Cell. Biol. 13 (3) (1993) 1440–1448.
[14] P.D. Carr, S.E. Gustin, A.P. Church, J.M. Murphy, S.C. Ford, D.A. Mann, D.M. Woltring, I. Walker, D.L. Ollis, I.G. Young, Structure of the complete extracellular domain of the common beta subunit of the human GM-CSF, IL-3, and IL-5 receptors reveals a novel dimer configuration, Cell 104 (2) (2001) 291–300.
[15] B.G. Moon, S. Takaki, K. Miyake, K. Takatsu, The role of IL-5 for mature B-1 cells in homeostatic proliferation, cell survival, and Ig production, J. Immunol. 172 (10) (2004) 6020–6029.
[16] K. Horikawa, K. Takatsu, Interleukin-5 regulates genes involved in B-cell terminal maturation, Immunology 118 (4) (2006) 497–508.
[17] C. Mizoguchi, S. Uehara, S. Akira, K. Takatsu, IL-5 induces IgG1 isotype switch recombination in mouse CD38-activated sIgD-positive B lymphocytes, J. Immunol. 162 (5) (1999) 2812–2819.
[18] K. Horikawa, H. Kaku, H. Nakajima, H.W. Davey, L. Hennighausen, I. Iwamoto, T. Yasue, A. Kariyone, K. Takatsu, Essential role of Stat5 for IL-5-dependent IgH switch recombination in mouse B cells, J. Immunol. 167 (9) (2001) 5018–5026.
[19] M. Plaut, J.H. Pierce, C.J. Watson, J. Hanley-Hyde, R.P. Nordan, W.E. Paul, Mast cell lines produce lymphokines in response to cross-linkage of Fc epsilon RI or to calcium ionophores, Nature 339 (6219) (1989) 64–67.
[20] M. Ikutani, T. Yanagibashi, M. Ogasawara, K. Tsuneyama, S. Yamamoto, Y. Hattori, T. Kouro, A. Itakura, Y. Nagai, S. Takaki, K. Takatsu, Identification of innate IL-5-producing cells and their role in lung eosinophil regulation and antitumor immunity, J. Immunol. 188 (2) (2012) 703–713.
[21] C.S. Klose, D. Artis, Innate lymphoid cells as regulators of immunity, inflammation and tissue homeostasis, Nat. Immunol. 17 (7) (2016) 765–774.
[22] S.P. Hogan, H.F. Rosenberg, R. Moqbel, S. Phipps, P.S. Foster, P. Lacy, A.B. Kay, M.E. Rothenberg, Eosinophils: biological properties and role in health and disease, Clin. Exp. Allergy 38 (5) (2008) 709–750.
[23] H. Tanaka, N. Kawada, T. Yamada, K. Kawada, K. Takatsu, H. Nagai, Allergen-induced airway inflammation and bronchial responsiveness in interleukin-5 receptor alpha chain-deficient mice, Clin. Exp. Allergy 30 (6) (2000) 874–881.
[24] I. Akutsu, T. Kojima, A. Kariyone, T. Fukuda, S. Makino, K. Takatsu, Antibody against interleukin-5 prevents antigen-induced eosinophil infiltration and bronchial hyperreactivity in the guinea pig airways, Immunol. Lett. 45 (1–2) (1995) 109–116.
[25] P. Nair, M.M. Pizzichini, M. Kjarsgaard, M.D. Inman, A. Efthimiadis, E. Pizzichini, F.E. Hargreave, P.M. O'Byrne, Mepolizumab for prednisone-dependent asthma with sputum eosinophilia, N. Engl. J. Med. 360 (10) (2009) 985–993.
[26] P. Haldar, C.E. Brightling, B. Hargadon, S. Gupta, W. Monteiro, A. Sousa, R.P. Marshall, P. Bradding, R.H. Green, A.J. Wardlaw, I.D. Pavord, Mepolizumab and exacerbations of refractory eosinophilic asthma, N. Engl. J. Med. 360 (10) (2009) 973–984.
[27] J.C. Kips, B.J. O'Connor, S.J. Langley, A. Woodcock, H.A. Kerstjens, D.S. Postma, M. Danzig, F. Cuss, R.A. Pauwels, Effect of SCH55700, a humanized anti-human interleukin-5 antibody, in severe persistent asthma: a pilot study, Am. J. Respir. Crit. Care Med. 167 (12) (2003) 1655–1659.
[28] K. Takatsu, K. Tanaka, A. Tominaga, Y. Kumahara, T. Hamaoka, Antigen-induced T cell-replacing factor (TRF). III. Establishment of T cell hybrid clone continuously producing TRF and functional analysis of released TRF, J. Immunol. 125 (6) (1980) 2646–2653.
[29] N. Harada, Y. Kikuchi, A. Tominaga, S. Takaki, K. Takatsu, BCGFII activity on activated B cells of a purified murine T cell-replacing factor (TRF) from a T cell hybridoma (B151K12), J. Immunol. 134 (6) (1985) 3944–3951.
[30] K. Takatsu, Y. Kikuchi, T. Takahashi, T. Honjo, M. Matsumoto, N. Harada, N. Yamaguchi, A. Tominaga, Interleukin 5, a T-cell-derived B-cell differentiation factor also induces cytotoxic T lymphocytes, Proc. Natl. Acad. Sci. USA 84 (12) (1987) 4234–4238.
[31] Y. Yamaguchi, T. Suda, J. Suda, M. Eguchi, Y. Miura, N. Harada, A. Tominaga, K. Takatsu, Purified interleukin 5 supports the terminal differentiation and proliferation of murine eosinophilic precursors, J. Exp. Med. 167 (1) (1988) 43–56.
[32] A.F. Lopez, C.G. Begley, D.J. Williamson, D.J. Warren, M.A. Vadas, C.J. Sanderson, Murine eosinophil differentiation factor. An eosinophil-specific colony-stimulating factor with activity for human cells, J. Exp. Med. 163 (5) (1986) 1085–1099.
[33] C. Azuma, T. Tanabe, M. Konishi, T. Kinashi, T. Noma, F. Matsuda, Y. Yaoita, K. Takatsu, L. Hammarstrom, C.I. Smith, et al., Cloning of cDNA for human T-cell replacing factor (interleukin-5) and comparison with the murine homologue, Nucl. Acids Res. 14 (22) (1986) 9149–9158.
[34] K. Takatsu, S. Takaki, Y. Hitoshi, Interleukin-5 and its receptor system: implications in the immune system and inflammation, Adv. Immunol. 57 (1994) 145–190.
[35] A. Tominaga, T. Takahashi, Y. Kikuchi, S. Mita, S. Naomi, N. Harada, N. Yamaguchi, K. Takatsu, Role of carbohydrate moiety of IL-5. Effect of tunicamycin on the glycosylation of IL-5 and the biologic activity of deglycosylated IL-5, J. Immunol. 144 (4) (1990) 1345–1352.
[36] M.V. Milburn, A.M. Hassell, M.H. Lambert, S.R. Jordan, A.E. Proudfoot, P. Graber, T.N. Wells, A novel dimer configuration revealed by the crystal structure at 2.4 A resolution of human interleukin-5, Nature 363 (6425) (1993) 172–176.
[37] S. Mita, N. Harada, S. Naomi, Y. Hitoshi, K. Sakamoto, M. Akagi, A. Tominaga, K. Takatsu, Receptors for T cell-replacing factor/interleukin 5. Specificity, quantitation, and its implication, J. Exp. Med. 168 (3) (1988) 863–878.
[38] S. Mita, A. Tominaga, Y. Hitoshi, K. Sakamoto, T. Honjo, M. Akagi, Y. Kikuchi, N. Yamaguchi, K. Takatsu, Characterization of high-affinity receptors for interleukin 5 on interleukin 5-dependent cell lines, Proc. Natl. Acad. Sci. USA 86 (7) (1989) 2311–2315.
[39] S. Mita, S. Takaki, Y. Hitoshi, A.G. Rolink, A. Tominaga, N. Yamaguchi, K. Takatsu, Molecular characterization of the beta chain of the murine interleukin 5 receptor, Int. Immunol. 3 (7) (1991) 665–672.
[40] Y. Hitoshi, N. Yamaguchi, S. Mita, E. Sonoda, S. Takaki, A. Tominaga, K. Takatsu, Distribution of IL-5 receptor-positive B cells. Expression of IL-5 receptor on Ly-1(CD5)+ B cells, J. Immunol. 144 (11) (1990) 4218–4225.
[41] S. Takaki, H. Kanazawa, M. Shiiba, K. Takatsu, A critical cytoplasmic domain of the interleukin-5 (IL-5) receptor alpha chain and its function in IL-5-mediated growth signal transduction, Mol. Cell. Biol. 14 (11) (1994) 7404–7413.
[42] T. Kouro, Y. Kikuchi, H. Kanazawa, K. Hirokawa, N. Harada, M. Shiiba, H. Wakao, S. Takaki, K. Takatsu, Critical proline residues of the cytoplasmic domain of the IL-5 receptor alpha chain and its function in IL-5-mediated activation of JAK kinase and STAT5, Int. Immunol. 8 (2) (1996) 237–245.
[43] B.G. Moon, T. Yoshida, M. Shiiba, K. Nakao, M. Katsuki, S. Takaki, K. Takatsu, Functional dissection of the cytoplasmic subregions of the interleukin-5

receptor alpha chain in growth and immunoglobulin G1 switch recombination of B cells, Immunology 102 (3) (2001) 289–300.
[44] S. Kusano, M. Kukimoto-Niino, N. Hino, N. Ohsawa, M. Ikutani, S. Takaki, K. Sakamoto, M. Hara-Yokoyama, M. Shirouzu, K. Takatsu, S. Yokoyama, Structural basis of interleukin-5 dimer recognition by its alpha receptor, Protein Sci. 21 (6) (2012) 850–864.
[45] E. Patino, A. Kotzsch, S. Saremba, J. Nickel, W. Schmitz, W. Sebald, T.D. Mueller, Structure analysis of the IL-5 ligand-receptor complex reveals a wrench-like architecture for IL-5Ralpha, Structure 19 (12) (2011) 1864–1875.
[46] N. Ogata, T. Kouro, A. Yamada, M. Koike, N. Hanai, T. Ishikawa, K. Takatsu, JAK2 and JAK1 constitutively associate with an interleukin-5 (IL-5) receptor alpha and betac subunit, respectively, and are activated upon IL-5 stimulation, Blood 91 (7) (1998) 2264–2271.
[47] S. Kagami, H. Nakajima, A. Suto, K. Hirose, K. Suzuki, S. Morita, I. Kato, Y. Saito, T. Kitamura, I. Iwamoto, Stat5a regulates T helper cell differentiation by several distinct mechanisms, Blood 97 (8) (2001) 2358–2365.
[48] S. Sato, T. Katagiri, S. Takaki, Y. Kikuchi, Y. Hitoshi, S. Yonehara, S. Tsukada, D. Kitamura, T. Watanabe, O. Witte, K. Takatsu, IL-5 receptor-mediated tyrosine phosphorylation of SH2/SH3-containing proteins and activation of Bruton's tyrosine and Janus 2 kinases, J. Exp. Med. 180 (6) (1994) 2101–2111.
[49] K. Takatsu, Interleukin-5, Curr. Opin. Immunol. 4 (3) (1992) 299–306.
[50] K. Pazdrak, D. Schreiber, P. Forsythe, L. Justement, R. Alam, The intracellular signal transduction mechanism of interleukin 5 in eosinophils: the involvement of lyn tyrosine kinase and the Ras-Raf-1-MEK-microtubule-associated protein kinase pathway, J. Exp. Med. 181 (5) (1995) 1827–1834.
[51] D.J. Hall, J. Cui, M.E. Bates, B.A. Stout, L. Koenderman, P.J. Coffer, P.J. Bertics, Transduction of a dominant-negative H-Ras into human eosinophils attenuates extracellular signal-regulated kinase activation and interleukin-5-mediated cell viability, Blood 98 (7) (2001) 2014–2021.
[52] K.W. Beagley, J.H. Eldridge, H. Kiyono, M.P. Everson, W.J. Koopman, T. Honjo, J. R. McGhee, Recombinant murine IL-5 induces high rate IgA synthesis in cycling IgA-positive Peyer's patch B cells, J. Immunol. 141 (6) (1988) 2035–2042.
[53] R.L. Coffman, B. Shrader, J. Carty, T.R. Mosmann, M.W. Bond, A mouse T cell product that preferentially enhances IgA production. I. Biologic characterization, J. Immunol. 139 (11) (1987) 3685–3690.
[54] G.R. Harriman, D.Y. Kunimoto, J.F. Elliott, V. Paetkau, W. Strober, The role of IL-5 in IgA B cell differentiation, J. Immunol. 140 (9) (1988) 3033–3039.
[55] R. Matsumoto, M. Matsumoto, S. Mita, Y. Hitoshi, M. Ando, S. Araki, N. Yamaguchi, A. Tominaga, K. Takatsu, Interleukin-5 induces maturation but not class switching of surface IgA-positive B cells into IgA-secreting cells, Immunology 66 (1) (1989) 32–38.
[56] E. Sonoda, R. Matsumoto, Y. Hitoshi, T. Ishii, M. Sugimoto, S. Araki, A. Tominaga, N. Yamaguchi, K. Takatsu, Transforming growth factor beta induces IgA production and acts additively with interleukin 5 for IgA production, J. Exp. Med. 170 (4) (1989) 1415–1420.
[57] T. Hiroi, M. Yanagita, H. Iijima, K. Iwatani, T. Yoshida, K. Takatsu, H. Kiyono, Deficiency of IL-5 receptor alpha-chain selectively influences the development of the common mucosal immune system independent IgA-producing B-1 cell in mucosa-associated tissues, J. Immunol. 162 (2) (1999) 821–828.
[58] N. Harada, M. Matsumoto, N. Koyama, A. Shimizu, T. Honjo, A. Tominaga, K. Takatsu, T cell replacing factor/interleukin 5 induces not only B-cell growth and differentiation, but also increased expression of interleukin 2 receptor on activated B-cells, Immunol. Lett. 15 (3) (1987) 205–215.
[59] M.S. Loughnan, K. Takatsu, N. Harada, G.J. Nossal, T-cell-replacing factor (interleukin 5) induces expression of interleukin 2 receptors on murine splenic B cells, Proc. Natl. Acad. Sci. USA 84 (15) (1987) 5399–5403.
[60] K. Nakanishi, T. Hashimoto, K. Hiroishi, K. Matsui, T. Yoshimoto, H.C. Morse 3rd, J. Furuyama, T. Hamaoka, K. Higashino, W.E. Paul, Demonstration of up-regulated IL 2 receptor expression on an in vitro cloned BCL1 subline, J. Immunol. 138 (6) (1987) 1817–1825.
[61] Y. Hitoshi, E. Sonoda, Y. Kikuchi, S. Yonehara, H. Nakauchi, K. Takatsu, IL-5 receptor positive B cells, but not eosinophils, are functionally and numerically influenced in mice carrying the X-linked immune defect, Int. Immunol. 5 (9) (1993) 1183–1190.
[62] T. Yoshida, K. Ikuta, H. Sugaya, K. Maki, M. Takagi, H. Kanazawa, S. Sunaga, T. Kinashi, K. Yoshimura, J. Miyazaki, S. Takaki, K. Takatsu, Defective B-1 cell development and impaired immunity against Angiostrongylus cantonensis in IL-5R alpha-deficient mice, Immunity 4 (5) (1996) 483–494.
[63] K. Takatsu, Interleukin 5 and B cell differentiation, Cytokine Growth Factor Rev. 9 (1) (1998) 25–35.
[64] R.I. Tepper, R.L. Coffman, P. Leder, An eosinophil-dependent mechanism for the antitumor effect of interleukin-4, Science 257 (5069) (1992) 548–551.
[65] R.I. Tepper, P.K. Pattengale, P. Leder, Murine interleukin-4 displays potent anti-tumor activity in vivo, Cell 57 (3) (1989) 503–512.
[66] V.T. Chu, A. Frohlich, G. Steinhauser, T. Scheel, T. Roch, S. Fillatreau, J.J. Lee, M. Lohning, C. Berek, Eosinophils are required for the maintenance of plasma cells in the bone marrow, Nat. Immunol. 12 (2) (2011) 151–159.
[67] V.T. Chu, A. Beller, S. Rausch, J. Strandmark, M. Zanker, O. Arbach, A. Kruglov, C. Berek, Eosinophils promote generation and maintenance of immunoglobulin-A-expressing plasma cells and contribute to gut immune homeostasis, Immunity 40 (4) (2014) 582–593.
[68] J.J. Lee, D. Dimina, M.P. Macias, S.I. Ochkur, M.P. McGarry, K.R. O'Neill, C. Protheroe, R. Pero, T. Nguyen, S.A. Cormier, E. Lenkiewicz, D. Colbert, L. Rinaldi, S.J. Ackerman, C.G. Irvin, N.A. Lee, Defining a link with asthma in mice congenitally deficient in eosinophils, Science 305 (5691) (2004) 1773–1776.
[69] C. Yu, A.B. Cantor, H. Yang, C. Browne, R.A. Wells, Y. Fujiwara, S.H. Orkin, Targeted deletion of a high-affinity GATA-binding site in the GATA-1 promoter leads to selective loss of the eosinophil lineage in vivo, J. Exp. Med. 195 (11) (2002) 1387–1395.
[70] A.A. Humbles, C.M. Lloyd, S.J. McMillan, D.S. Friend, G. Xanthou, E.E. McKenna, S. Ghiran, N.P. Gerard, C. Yu, S.H. Orkin, C. Gerard, A critical role for eosinophils in allergic airways remodeling, Science 305 (5691) (2004) 1776–1779.
[71] A. Tominaga, S. Takaki, N. Koyama, S. Katoh, R. Matsumoto, M. Migita, Y. Hitoshi, Y. Hosoya, S. Yamauchi, Y. Kanai, et al., Transgenic mice expressing a B cell growth and differentiation factor gene (interleukin 5) develop eosinophilia and autoantibody production, J. Exp. Med. 173 (2) (1991) 429–437.
[72] L.A. Dent, M. Strath, A.L. Mellor, C.J. Sanderson, Eosinophilia in transgenic mice expressing interleukin 5, J. Exp. Med. 172 (5) (1990) 1425–1431.
[73] H. Nakajima, I. Iwamoto, S. Tomoe, R. Matsumura, H. Tomioka, K. Takatsu, S. Yoshida, CD4+ T-lymphocytes and interleukin-5 mediate antigen-induced eosinophil infiltration into the mouse trachea, Am. Rev. Respir. Dis. 146 (2) (1992) 374–377.
[74] A.J. Van Oosterhout, A.R. Ladenius, H.F. Savelkoul, I. Van Ark, K.C. Delsman, F. P. Nijkamp, Effect of anti-IL-5 and IL-5 on airway hyperreactivity and eosinophils in guinea pigs, Am. Rev. Respir. Dis. 147 (3) (1993) 548–552.
[75] P.J. Mauser, A.M. Pitman, X. Fernandez, S.K. Foran, G.K. Adams 3rd, W. Kreutner, R.W. Egan, R.W. Chapman, Effects of an antibody to interleukin-5 in a monkey model of asthma, Am. J. Respir. Crit. Care Med. 152 (2) (1995) 467–472.
[76] P.S. Foster, S.P. Hogan, A.J. Ramsay, K.I. Matthaei, I.G. Young, Interleukin 5 deficiency abolishes eosinophilia, airways hyperreactivity, and lung damage in a mouse asthma model, J. Exp. Med. 183 (1) (1996) 195–201.
[77] M. Kopf, F. Brombacher, P.D. Hodgkin, A.J. Ramsay, E.A. Milbourne, W.J. Dai, K. S. Ovington, C.A. Behm, G. Kohler, I.G. Young, K.I. Matthaei, IL-5-deficient mice have a developmental defect in CD5+ B-1 cells and lack eosinophilia but have normal antibody and cytotoxic T cell responses, Immunity 4 (1) (1996) 15–24.
[78] P.G. Fallon, S.J. Ballantyne, N.E. Mangan, J.L. Barlow, A. Dasvarma, D.R. Hewett, A. McIlgorm, H.E. Jolin, A.N. McKenzie, 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. 203 (4) (2006) 1105–1116.
[79] M.M. Fort, J. Cheung, D. Yen, J. Li, S.M. Zurawski, S. Lo, S. Menon, T. Clifford, B. Hunte, R. Lesley, T. Muchamuel, S.D. Hurst, G. Zurawski, M.W. Leach, D.M. Gorman, D.M. Rennick, IL-25 induces IL-4, IL-5, and IL-13 and Th2-associated pathologies in vivo, Immunity 15 (6) (2001) 985–995.
[80] D.R. Neill, S.H. Wong, A. Bellosi, R.J. Flynn, M. Daly, T.K. Langford, C. Bucks, C. M. Kane, P.G. Fallon, R. Pannell, H.E. Jolin, A.N. McKenzie, Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity, Nature 464 (7293) (2010) 1367–1370.
[81] A.E. Price, H.E. Liang, B.M. Sullivan, R.L. Reinhardt, C.J. Eisley, D.J. Erle, R.M. Locksley, Systemically dispersed innate IL-13-expressing cells in type 2 immunity, Proc. Natl. Acad. Sci. USA 107 (25) (2010) 11489–11494.
[82] K. Moro, T. Yamada, M. Tanabe, T. Takeuchi, T. Ikawa, H. Kawamoto, J. Furusawa, M. Ohtani, H. Fujii, S. Koyasu, Innate production of T(H)2 cytokines by adipose tissue-associated c-Kit(+)Sca-1(+) lymphoid cells, Nature 463 (7280) (2010) 540–544.
[83] H. Spits, D. Artis, M. Colonna, A. Diefenbach, J.P. Di Santo, G. Eberl, S. Koyasu, R.M. Locksley, A.N. McKenzie, R.E. Mebius, F. Powrie, E. Vivier, Innate lymphoid cells–a proposal for uniform nomenclature, Nat. Rev. Immunol. 13 (2) (2013) 145–149.
[84] W.E. Paul, J. Zhu, How are T(H)2-type immune responses initiated and amplified?, Nat Rev. Immunol. 10 (4) (2010) 225–235.
[85] J.L. Barlow, A. Bellosi, C.S. Hardman, L.F. Drynan, S.H. Wong, J.P. Cruickshank, A.N. McKenzie, Innate IL-13-producing nuocytes arise during allergic lung inflammation and contribute to airways hyperreactivity, J. Allergy Clin. Immunol. 129 (1) (2012). 191–198.e4.
[86] D.M. Walter, J.J. McIntire, G. Berry, A.N. McKenzie, D.D. Donaldson, R.H. DeKruyff, D.T. Umetsu, Critical role for IL-13 in the development of allergen-induced airway hyperreactivity, J. Immunol. 167 (8) (2001) 4668–4675.
[87] C.S. Klose, M. Flach, L. Mohle, L. Rogell, T. Hoyler, K. Ebert, C. Fabiunke, D. Pfeifer, V. Sexl, D. Fonseca-Pereira, R.G. Domingues, H. Veiga-Fernandes, S.J. Arnold, M. Busslinger, I.R. Dunay, Y. Tanriver, A. Diefenbach, Differentiation of type 1 ILCs from a common progenitor to all helper-like innate lymphoid cell lineages, Cell 157 (2) (2014) 340–356.
[88] T. Hoyler, C.S. Klose, A. Souabni, A. Turqueti-Neves, D. Pfeifer, E.L. Rawlins, D. Voehringer, M. Busslinger, A. Diefenbach, The transcription factor GATA-3 controls cell fate and maintenance of type 2 innate lymphoid cells, Immunity 37 (4) (2012) 634–648.
[89] T.Y. Halim, A. MacLaren, M.T. Romanish, M.J. Gold, K.M. McNagny, F. Takei, Retinoic-acid-receptor-related orphan nuclear receptor alpha is required for natural helper cell development and allergic inflammation, Immunity 37 (3) (2012) 463–474.
[90] S.H. Wong, J.A. Walker, H.E. Jolin, L.F. Drynan, E. Hams, A. Camelo, J.L. Barlow, D.R. Neill, V. Panova, U. Koch, F. Radtke, C.S. Hardman, Y.Y. Hwang, P.G. Fallon, A.N. McKenzie, Transcription factor RORalpha is critical for nuocyte development, Nat. Immunol. 13 (3) (2012) 229–236.

[91] C.J. Spooner, J. Lesch, D. Yan, A.A. Khan, A. Abbas, V. Ramirez-Carrozzi, M. Zhou, R. Soriano, J. Eastham-Anderson, L. Diehl, W.P. Lee, Z. Modrusan, R. Pappu, M. Xu, J. DeVoss, H. Singh, Specification of type 2 innate lymphocytes by the transcriptional determinant Gfi1, Nat. Immunol. 14 (12) (2013) 1229–1236.
[92] J. Mjosberg, J. Bernink, K. Golebski, J.J. Karrich, C.P. Peters, B. Blom, A.A. te Velde, W.J. Fokkens, C.M. van Drunen, H. Spits, The transcription factor GATA3 is essential for the function of human type 2 innate lymphoid cells, Immunity 37 (4) (2012) 649–659.
[93] Q. Yang, L.A. Monticelli, S.A. Saenz, A.W. Chi, G.F. Sonnenberg, J. Tang, M.E. De Obaldia, W. Bailis, J.L. Bryson, K. Toscano, J. Huang, A. Haczku, W.S. Pear, D. Artis, A. Bhandoola, T cell factor 1 is required for group 2 innate lymphoid cell generation, Immunity 38 (4) (2013) 694–704.
[94] Y. Yu, C. Wang, S. Clare, J. Wang, S.C. Lee, C. Brandt, S. Burke, L. Lu, D. He, N.A. Jenkins, N.G. Copeland, G. Dougan, P. Liu, The transcription factor Bcl11b is specifically expressed in group 2 innate lymphoid cells and is essential for their development, J. Exp. Med. 212 (6) (2015) 865–874.
[95] J.A. Walker, C.J. Oliphant, A. Englezakis, Y. Yu, S. Clare, H.R. Rodewald, G. Belz, P. Liu, P.G. Fallon, A.N. McKenzie, Bcl11b is essential for group 2 innate lymphoid cell development, J. Exp. Med. 212 (6) (2015) 875–882.
[96] R.J. Noelle, E.C. Nowak, Cellular sources and immune functions of interleukin-9, Nat. Rev. Immunol. 10 (10) (2010) 683–687.
[97] J.E. Turner, P.J. Morrison, C. Wilhelm, M. Wilson, H. Ahlfors, J.C. Renauld, U. Panzer, H. Helmby, B. Stockinger, IL-9-mediated survival of type 2 innate lymphoid cells promotes damage control in helminth-induced lung inflammation, J. Exp. Med. 210 (13) (2013) 2951–2965.
[98] C. Wilhelm, K. Hirota, B. Stieglitz, J. Van Snick, M. Tolaini, K. Lahl, T. Sparwasser, H. Helmby, B. Stockinger, An IL-9 fate reporter demonstrates the induction of an innate IL-9 response in lung inflammation, Nat. Immunol. 12 (11) (2011) 1071–1077.
[99] P. Licona-Limon, J. Henao-Mejia, A.U. Temann, N. Gagliani, I. Licona-Limon, H. Ishigame, L. Hao, D.R. Herbert, R.A. Flavell, Th9 cells drive host immunity against gastrointestinal worm infection, Immunity 39 (4) (2013) 744–757.
[100] A. Mohapatra, S.J. Van Dyken, C. Schneider, J.C. Nussbaum, H.E. Liang, R.M. Locksley, Group 2 innate lymphoid cells utilize the IRF4-IL-9 module to coordinate epithelial cell maintenance of lung homeostasis, Mucosal Immunol. 9 (1) (2016) 275–286.
[101] M. Veldhoen, V. Brucklacher-Waldert, Dietary influences on intestinal immunity, Nat. Rev. Immunol. 12 (10) (2012) 696–708.
[102] S.P. Spencer, C. Wilhelm, Q. Yang, J.A. Hall, N. Bouladoux, A. Boyd, T.B. Nutman, J.F. Urban Jr., J. Wang, T.R. Ramalingam, A. Bhandoola, T.A. Wynn, Y. Belkaid, Adaptation of innate lymphoid cells to a micronutrient deficiency promotes type 2 barrier immunity, Science 343 (6169) (2014) 432–437.
[103] J.C. Nussbaum, S.J. Van Dyken, J. von Moltke, L.E. Cheng, A. Mohapatra, A.B. Molofsky, E.E. Thornton, M.F. Krummel, A. Chawla, H.E. Liang, R.M. Locksley, Type 2 innate lymphoid cells control eosinophil homeostasis, Nature 502 (7470) (2013) 245–248.
[104] K. Moro, H. Kabata, M. Tanabe, S. Koga, N. Takeno, M. Mochizuki, K. Fukunaga, K. Asano, T. Betsuyaku, S. Koyasu, Interferon and IL-27 antagonize the function of group 2 innate lymphoid cells and type 2 innate immune responses, Nat. Immunol. 17 (1) (2016) 76–86.
[105] C.U. Duerr, C.D. McCarthy, B.C. Mindt, M. Rubio, A.P. Meli, J. Pothlichet, M.M. Eva, J.F. Gauchat, S.T. Qureshi, B.D. Mazer, K.L. Mossman, D. Malo, A.M. Gamero, S.M. Vidal, I.L. King, M. Sarfati, J.H. Fritz, Type I interferon restricts type 2 immunopathology through the regulation of group 2 innate lymphoid cells, Nat. Immunol. 17 (1) (2016) 65–75.
[106] F. Kudo, M. Ikutani, Y. Seki, T. Otsubo, Y.I. Kawamura, T. Dohi, K. Oshima, M. Hattori, S. Nakae, K. Takatsu, S. Takaki, Interferon-gamma constrains cytokine production of group 2 innate lymphoid cells, Immunology 147 (1) (2016) 21–29.
[107] N. Krishnamoorthy, P.R. Burkett, J. Dalli, R.E. Abdulnour, R. Colas, S. Ramon, R. P. Phipps, N.A. Petasis, V.K. Kuchroo, C.N. Serhan, B.D. Levy, Cutting edge: maresin-1 engages regulatory T cells to limit type 2 innate lymphoid cell activation and promote resolution of lung inflammation, J. Immunol. 194 (3) (2015) 863–867.
[108] C. Barnig, B.D. Levy, Innate immunity is a key factor for the resolution of inflammation in asthma, Eur. Respir. Rev. 24 (135) (2015) 141–153.
[109] M. Bemark, P. Boysen, N.Y. Lycke, Induction of gut IgA production through T cell-dependent and T cell-independent pathways, Ann. NY Acad. Sci. 1247 (2012) 97–116.
[110] T.W. Wong, A.D. Doyle, J.J. Lee, D.F. Jelinek, Eosinophils regulate peripheral B cell numbers in both mice and humans, J. Immunol. 192 (8) (2014) 3548–3558.
[111] C.M. Lloyd, E.M. Hessel, Functions of T cells in asthma: more than just T(H)2 cells, Nat. Rev. Immunol. 10 (12) (2010) 838–848.
[112] H.J. Gould, B.J. Sutton, IgE in allergy and asthma today, Nat. Rev. Immunol. 8 (3) (2008) 205–217.
[113] M. Wills-Karp, Interleukin-13 in asthma pathogenesis, Immunol. Rev. 202 (2004) 175–190.
[114] J.R. Rock, B.L. Hogan, Epithelial progenitor cells in lung development, maintenance, repair, and disease, Annu. Rev. Cell Dev. Biol. 27 (2011) 493–512.
[115] T.Y. Halim, R.H. Krauss, A.C. Sun, F. Takei, Lung natural helper cells are a critical source of Th2 cell-type cytokines in protease allergen-induced airway inflammation, Immunity 36 (3) (2012) 451–463.
[116] K.R. Bartemes, K. Iijima, T. Kobayashi, G.M. Kephart, A.N. McKenzie, H. Kita, IL-33-responsive lineage- CD25+ CD44(hi) lymphoid cells mediate innate type 2 immunity and allergic inflammation in the lungs, J. Immunol. 188 (3) (2012) 1503–1513.
[117] D.E. Byers, M.J. Holtzman, Alternatively activated macrophages and airway disease, Chest 140 (3) (2011) 768–774.
[118] S.J. Van Dyken, A. Mohapatra, J.C. Nussbaum, A.B. Molofsky, E.E. Thornton, S.F. Ziegler, A.N. McKenzie, M.F. Krummel, H.E. Liang, R.M. Locksley, Chitin activates parallel immune modules that direct distinct inflammatory responses via innate lymphoid type 2 and gammadelta T cells, Immunity 40 (3) (2014) 414–424.
[119] B.S. Kim, K. Wang, M.C. Siracusa, S.A. Saenz, J.R. Brestoff, L.A. Monticelli, M. Noti, E.D. Tait Wojno, T.C. Fung, M. Kubo, D. Artis, Basophils promote innate lymphoid cell responses in inflamed skin, J. Immunol. 193 (7) (2014) 3717–3725.
[120] S.G. Smith, R. Chen, M. Kjarsgaard, C. Huang, J.P. Oliveria, P.M. O'Byrne, G.M. Gauvreau, L.P. Boulet, C. Lemiere, J. Martin, P. Nair, R. Sehmi, Increased numbers of activated group 2 innate lymphoid cells in the airways of patients with severe asthma and persistent airway eosinophilia, J. Allergy Clin. Immunol. 137 (1) (2016). 75–86.e8.
[121] P. Nagakumar, L. Denney, L. Fleming, A. Bush, C.M. Lloyd, S. Saglani, Type 2 innate lymphoid cells in induced sputum from children with severe asthma, J. Allergy Clin. Immunol. 137 (2) (2016). 624–626.e6.
[122] K.R. Bartemes, G.M. Kephart, S.J. Fox, H. Kita, Enhanced innate type 2 immune response in peripheral blood from patients with asthma, J. Allergy Clin. Immunol. 134 (3) (2014). 671–678.e4.
[123] C. Ober, T.C. Yao, The genetics of asthma and allergic disease: a 21st century perspective, Immunol. Rev. 242 (1) (2011) 10–30.
[124] D.G. Torgerson, E.J. Ampleford, G.Y. Chiu, W.J. Gauderman, C.R. Gignoux, P.E. Graves, B.E. Himes, A.M. Levin, R.A. Mathias, D.B. Hancock, J.W. Baurley, C. Eng, D.A. Stern, J.C. Celedon, N. Rafaels, D. Capurso, D.V. Conti, L.A. Roth, M. Soto-Quiros, A. Togias, X. Li, R.A. Myers, I. Romieu, D.J. Van Den Berg, D. Hu, N. N. Hansel, R.D. Hernandez, E. Israel, M.T. Salam, J. Galanter, P.C. Avila, L. Avila, J.R. Rodriquez-Santana, R. Chapela, W. Rodriguez-Cintron, G.B. Diette, N.F. Adkinson, R.A. Abel, K.D. Ross, M. Shi, M.U. Faruque, G.M. Dunston, H.R. Watson, V.J. Mantese, S.C. Ezurum, L. Liang, I. Ruczinski, J.G. Ford, S. Huntsman, K.F. Chung, H. Vora, X. Li, W.J. Calhoun, M. Castro, J.J. Sienra-Monge, B. del Rio-Navarro, K.A. Deichmann, A. Heinzmann, S.E. Wenzel, W.W. Busse, J.E. Gern, R.F. Lemanske Jr., T.H. Beaty, E.R. Bleecker, B.A. Raby, D.A. Meyers, S.J. LondonS. Mexico City Childhood Asthma, F.D. GillilandS. Children's Health, H. study, E.G. BurchardS.o.G.E. Genetics of Asthma in Latino Americans Study, A. Admixture in Latino, A.G. Study of African Americans, Environments, F.D. MartinezR. Childhood Asthma, N. Education, S. T. WeissP. Childhood Asthma Management, L.K. WilliamsP. Study of Asthma, R.-E. Pharmacogenomic Interactions, K.C. BarnesS. Genetic Research on Asthma in African Diaspora, C. Ober, D.L. Nicolae, Meta-analysis of genome-wide association studies of asthma in ethnically diverse North American populations, Nat. Genet. 43 (9) (2011) 887–892.
[125] J.S. Jung, B.L. Park, H.S. Cheong, J.S. Bae, J.H. Kim, H.S. Chang, T. Rhim, J.S. Park, A.S. Jang, Y.M. Lee, K.U. Kim, S.T. Uh, J.O. Na, Y.H. Kim, C.S. Park, H.D. Shin, Association of IL-17RB gene polymorphism with asthma, Chest 135 (5) (2009) 1173–1180.
[126] M.F. Moffatt, I.G. Gut, F. Demenais, D.P. Strachan, E. Bouzigon, S. Heath, E. von Mutius, M. Farrall, M. Lathrop, W.O. Cookson, G. Consortium, A large-scale, consortium-based genomewide association study of asthma, N. Engl. J. Med. 363 (13) (2010) 1211–1221.
[127] C.A. Wu, L. Puddington, H.E. Whiteley, C.A. Yiamouyiannis, C.M. Schramm, F. Mohammadu, R.S. Thrall, Murine cytomegalovirus infection alters Th1/Th2 cytokine expression, decreases airway eosinophilia, and enhances mucus production in allergic airway disease, J. Immunol. 167 (5) (2001) 2798–2807.
[128] R. Almansa, M. Sanchez-Garcia, A. Herrero, S. Calzada, V. Roig, J. Barbado, L. Rico, F. Bobillo, J.M. Eiros, V. Iglesias, R.O. de Lejarazu, J.F. Bermejo-Martin, Host response cytokine signatures in viral and nonviral acute exacerbations of chronic obstructive pulmonary disease, J. Interferon Cytokine Res. 31 (5) (2011) 409–413.
[129] L.M. Looi, Tumor-associated tissue eosinophilia in nasopharyngeal carcinoma. A pathologic study of 422 primary and 138 metastatic tumors, Cancer 59 (3) (1987) 466–470.
[130] R.G. Dorta, G. Landman, L.P. Kowalski, J.R. Lauris, M.R. Latorre, D.T. Oliveira, Tumour-associated tissue eosinophilia as a prognostic factor in oral squamous cell carcinomas, Histopathology 41 (2) (2002) 152–157.
[131] H.J. Nielsen, U. Hansen, I.J. Christensen, C.M. Reimert, N. Brunner, F. Moesgaard, Independent prognostic value of eosinophil and mast cell infiltration in colorectal cancer tissue, J. Pathol. 189 (4) (1999) 487–495.
[132] G. Carlomagno, G. Messalli, R.M. Melillo, A.A. Stanziola, C. Visciano, V. Mercurio, M. Imbriaco, S. Ghio, M. Sofia, D. Bonaduce, S. Fazio, Serum soluble ST2 and interleukin-33 levels in patients with pulmonary arterial hypertension, Int. J. Cardiol. 168 (2) (2013) 1545–1547.
[133] M. Weng, D.M. Baron, K.D. Bloch, A.D. Luster, J.J. Lee, B.D. Medoff, Eosinophils are necessary for pulmonary arterial remodeling in a mouse model of eosinophilic inflammation-induced pulmonary hypertension, Am. J. Physiol. Lung Cell. Mol. Physiol. 301 (6) (2011) L927–L936.
[134] K.R. Tormanen, L. Uller, C.G. Persson, J.S. Erjefalt, Allergen exposure of mouse airways evokes remodeling of both bronchi and large pulmonary vessels, Am. J. Respir. Crit. Care Med. 171 (1) (2005) 19–25.

[135] K. Rydell-Tormanen, J.R. Johnson, R. Fattouh, M. Jordana, J.S. Erjefalt, Induction of vascular remodeling in the lung by chronic house dust mite exposure, Am. J. Respir. Cell Mol. Biol. 39 (1) (2008) 61–67.
[136] E. Daley, C. Emson, C. Guignabert, R. de Waal Malefyt, J. Louten, V.P. Kurup, C. Hogaboam, L. Taraseviciene-Stewart, N.F. Voelkel, M. Rabinovitch, E. Grunig, G. Grunig, Pulmonary arterial remodeling induced by a Th2 immune response, J. Exp. Med. 205 (2) (2008) 361–372.
[137] H. Kita, Eosinophils: multifaceted biological properties and roles in health and disease, Immunol. Rev. 242 (1) (2011) 161–177.
[138] D.B. Price, A. Rigazio, J.D. Campbell, E.R. Bleecker, C.J. Corrigan, M. Thomas, S. E. Wenzel, A.M. Wilson, M.B. Small, G. Gopalan, V.L. Ashton, A. Burden, E.V. Hillyer, M. Kerkhof, I.D. Pavord, Blood eosinophil count and prospective annual asthma disease burden: a UK cohort study, Lancet Respir. Med. 3 (11) (2015) 849–858.
[139] S.S. Eng, M.L. DeFelice, The role and immunobiology of eosinophils in the respiratory system: a comprehensive review, Clin. Rev. Allergy Immunol. 50 (2) (2016) 140–158.
[140] P. Haldar, I.D. Pavord, D.E. Shaw, M.A. Berry, M. Thomas, C.E. Brightling, A.J. Wardlaw, R.H. Green, Cluster analysis and clinical asthma phenotypes, Am. J. Respir. Crit. Care Med. 178 (3) (2008) 218–224.
[141] D.Y. Leung, S.J. Szefler, Diagnosis and management of steroid-resistant asthma, Clin. Chest Med. 18 (3) (1997) 611–625.
[142] C. Miranda, A. Busacker, S. Balzar, J. Trudeau, S.E. Wenzel, Distinguishing severe asthma phenotypes: role of age at onset and eosinophilic inflammation, J. Allergy Clin. Immunol. 113 (1) (2004) 101–108.
[143] S.E. Wenzel, L.B. Schwartz, E.L. Langmack, J.L. Halliday, J.B. Trudeau, R.L. Gibbs, H.W. Chu, Evidence that severe asthma can be divided pathologically into two inflammatory subtypes with distinct physiologic and clinical characteristics, Am. J. Respir. Crit. Care Med. 160 (3) (1999) 1001–1008.
[144] A. ten Brinke, A.H. Zwinderman, P.J. Sterk, K.F. Rabe, E.H. Bel, Factors associated with persistent airflow limitation in severe asthma, Am. J. Respir. Crit. Care Med. 164 (5) (2001) 744–748.
[145] S. Saglani, D.N. Payne, J. Zhu, Z. Wang, A.G. Nicholson, A. Bush, P.K. Jeffery, Early detection of airway wall remodeling and eosinophilic inflammation in preschool wheezers, Am. J. Respir. Crit. Care Med. 176 (9) (2007) 858–864.
[146] G.J. Gleich, C.R. Adolphson, The eosinophilic leukocyte: structure and function, Adv. Immunol. 39 (1986) 177–253.
[147] T. Morokata, K. Ida, T. Yamada, Characterization of YM-90709 as a novel antagonist which inhibits the binding of interleukin-5 to interleukin-5 receptor, Int. Immunopharmacol. 2 (12) (2002) 1693–1702.
[148] B.P. England, P. Balasubramanian, I. Uings, S. Bethell, M.J. Chen, P.J. Schatz, Q. Yin, Y.F. Chen, E.A. Whitehorn, A. Tsavaler, C.L. Martens, R.W. Barrett, M. McKinnon, A potent dimeric peptide antagonist of interleukin-5 that binds two interleukin-5 receptor alpha chains, Proc. Natl. Acad. Sci. USA 97 (12) (2000) 6862–6867.
[149] R. Devos, G. Plaetinck, S. Cornelis, Y. Guisez, J. Van der Heyden, J. Tavernier, Interleukin-5 and its receptor: a drug target for eosinophilia associated with chronic allergic disease, J. Leukoc. Biol. 57 (6) (1995) 813–819.
[150] M. McKinnon, K. Page, I.J. Uings, M. Banks, D. Fattah, A.E. Proudfoot, P. Graber, C. Arod, R. Fish, T.N. Wells, R. Solari, An interleukin 5 mutant distinguishes between two functional responses in human eosinophils, J. Exp. Med. 186 (1) (1997) 121–129.
[151] M.J. Leckie, A. ten Brinke, J. Khan, Z. Diamant, B.J. O'Connor, C.M. Walls, A.K. Mathur, H.C. Cowley, K.F. Chung, R. Djukanovic, T.T. Hansel, S.T. Holgate, P.J. Sterk, P.J. Barnes, Effects of an interleukin-5 blocking monoclonal antibody on eosinophils, airway hyper-responsiveness, and the late asthmatic response, Lancet 356 (9248) (2000) 2144–2148.
[152] P.T. Flood-Page, A.N. Menzies-Gow, A.B. Kay, D.S. Robinson, Eosinophil's role remains uncertain as anti-interleukin-5 only partially depletes numbers in asthmatic airway, Am. J. Respir. Crit. Care Med. 167 (2) (2003) 199–204.
[153] L. Bjermer, C. Lemiere, J. Maspero, S. Weiss, J. Zangrilli, M. Germinaro, Reslizumab for inadequately controlled asthma with elevated blood eosinophil levels: a randomized phase 3 study, Chest 150 (4) (2016) 789–798.
[154] J. Corren, S. Weinstein, L. Janka, J. Zangrilli, M. Garin, Phase 3 study of reslizumab in patients with poorly controlled asthma: effects across a broad range of eosinophil counts, Chest 150 (4) (2016) 799–810.
[155] R. Kolbeck, A. Kozhich, M. Koike, L. Peng, C.K. Andersson, M.M. Damschroder, J.L. Reed, R. Woods, W.W. Dall'acqua, G.L. Stephens, J.S. Erjefalt, L. Bjermer, A. A. Humbles, D. Gossage, H. Wu, P.A. Kiener, G.L. Spitalny, C.R. Mackay, N.A. Molfino, A.J. Coyle, MEDI-563, a humanized anti-IL-5 receptor alpha mAb with enhanced antibody-dependent cell-mediated cytotoxicity function, J. Allergy Clin. Immunol. 125 (6) (2010). 1344–1353.e2.
[156] L. George, C.E. Brightling, Eosinophilic airway inflammation: role in asthma and chronic obstructive pulmonary disease, Ther. Adv. Chronic Dis. 7 (1) (2016) 34–51.
[157] M. Koike, K. Nakamura, A. Furuya, A. Iida, H. Anazawa, K. Takatsu, N. Hanai, Establishment of humanized anti-interleukin-5 receptor alpha chain monoclonal antibodies having a potent neutralizing activity, Hum. Antibodies 18 (1–2) (2009) 17–27.
[158] W.W. Busse, R. Katial, D. Gossage, S. Sari, B. Wang, R. Kolbeck, A.J. Coyle, M. Koike, G.L. Spitalny, P.A. Kiener, G.P. Geba, N.A. Molfino, Safety profile, pharmacokinetics, and biologic activity of MEDI-563, an anti-IL-5 receptor alpha antibody, in a phase I study of subjects with mild asthma, J. Allergy Clin. Immunol. 125 (6) (2010). 1237–1244.e2.
[159] J.M. FitzGerald, E.R. Bleecker, P. Nair, S. Korn, K. Ohta, M. Lommatzsch, G.T. Ferguson, W.W. Busse, P. Barker, S. Sproule, G. Gilmartin, V. Werkstrom, M. Aurivillius, M. GoldmanC.s. investigators, Benralizumab, an anti-interleukin-5 receptor alpha monoclonal antibody, as add-on treatment for patients with severe, uncontrolled, eosinophilic asthma (CALIMA): a randomised, double-blind, placebo-controlled phase 3 trial, Lancet (2016).
[160] E.R. Bleecker, J.M. FitzGerald, P. Chanez, A. Papi, S.F. Weinstein, P. Barker, S. Sproule, G. Gilmartin, M. Aurivillius, V. Werkstrom, M. GoldmanS.s. investigators, Efficacy and safety of benralizumab for patients with severe asthma uncontrolled with high-dosage inhaled corticosteroids and long-acting beta2-agonists (SIROCCO): a randomised, multicentre, placebo-controlled phase 3 trial, Lancet (2016).
[161] T.H. Pham, G. Damera, P. Newbold, K. Ranade, Reductions in eosinophil biomarkers by benralizumab in patients with asthma, Respir. Med. 111 (2016) 21–29.
[162] M. Laviolette, D.L. Gossage, G. Gauvreau, R. Leigh, R. Olivenstein, R. Katial, W. W. Busse, S. Wenzel, Y. Wu, V. Datta, R. Kolbeck, N.A. Molfino, Effects of benralizumab on airway eosinophils in asthmatic patients with sputum eosinophilia, J. Allergy Clin. Immunol. 132 (5) (2013). 1086–1096.e5.
[163] L.D. Tan, J.M. Bratt, D. Godor, S. Louie, N.J. Kenyon, Benralizumab: a unique IL-5 inhibitor for severe asthma, J. Asthma Allergy 9 (2016) 71–81.
[164] M. Khorasanizadeh, M. Eskian, A.H. Assa'ad, C.A. Camargo Jr., N. Rezaei, Efficacy and safety of benralizumab, a monoclonal antibody against IL-5Ralpha, in uncontrolled eosinophilic asthma, Int. Rev. Immunol. (2016).