

PROF. KOICHIRO ASANO (Orcid ID : 0000-0002-9044-3061)

PROF. MAYUMI TAMARI (Orcid ID : 0000-0002-5755-9177)

Article type : Review

Invited review article

Adult-onset eosinophilic airway diseases

Short title: Adult-onset eosinophilic airway diseases

Koichiro Asano¹, Shigeharu Ueki², Mayumi Tamari³, Yoshimasa Imoto⁴, Shigeharu Fujieda⁴, and Masami Taniguchi⁵

¹Division of Pulmonary Medicine, Department of Medicine, Tokai University School of Medicine, Kanagawa, Japan

²Department of General Internal Medicine and Clinical Laboratory Medicine, Akita University Graduate School of Medicine, Akita, Japan

³Division of Molecular Genetics, Research Center for Medical Science, The Jikei University School of Medicine, Tokyo, Japan

⁴Division of Otorhinolaryngology–Head & Neck Surgery, Department of Sensory and Locomotor Medicine, Faculty of Medical Science, University of Fukui, Fukui, Japan

⁵Center for Allergy and Immunology, Shonan Kamakura General Hospital, Kanagawa, Japan

Corresponding author

Koichiro Asano, MD

Division of Pulmonary Medicine, Department of Medicine

Tokai University School of Medicine

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version of Record](#). Please cite this article as [doi: 10.1111/ALL.14620](https://doi.org/10.1111/ALL.14620)

This article is protected by copyright. All rights reserved

143 Shimokasuya, Isehara, Kanagawa 259-1193, Japan

Phone +81-463-93-1121

Fax +81-463-93-0381

E-mail: ko-asano@tokai-u.jp

Author Contributions:

All the authors have contributed equally to the drafting of the manuscript, have agreed to be accountable for all aspects of the study, and have approved the final version of the manuscript.

Word Count: 4,016 words

Acknowledgments

The authors are grateful to Ms. Satomi Misawa for outstanding assistance in drawing the figure.

This study was partly funded by a Research Grant on Allergic Disease and Immunology from the Japan Agency for Medical Research and Development under grant number JP20ek0410055 (KA), the Mochida Memorial Foundation for Medical and Pharmaceutical Research (SU), the Japan Society for the Promotion of Science KAKENHI (20K08794 to SU), the scientific research fund of the Ministry of Health, Labour and Welfare, Japan (201913009A to MT), and a Health Labour Sciences Research Grant (H30-Nantitou(nan)-Ippan-016) (SF).

Abstract

Eosinophilic airway inflammation is one of the cardinal features of allergic airway diseases such as atopic asthma and allergic rhinitis. These childhood-onset conditions are mediated by allergen and allergen-specific IgE and often accompanied by other allergic diseases including food allergy and eczema. They can develop consecutively in the same patient, which is referred to as an allergic march. In contrast, some phenotypes of asthma, nonsteroidal anti-inflammatory drugs-exacerbated airway disease (N-ERD), chronic rhinosinusitis with nasal polyps (CRSwNP)/eosinophilic CRS, and allergic bronchopulmonary aspergillosis/mycosis (ABPA/ABPM) are adult-onset airway diseases, which are characterised by prominent peripheral blood eosinophilia. Most of these conditions, except for ABPA/ABPM, are non-atopic, and the coexistence of multiple diseases, including an adult-onset eosinophilic systemic disease, eosinophilic granulomatosis with polyangiitis (EGPA), is common. In this review, we focus on eosinophil biology, genetics, and clinical characteristics and the pathophysiology of adult-onset eosinophilic asthma, N-ERD, CRSwNP/eosinophilic CRS, ABPA/ABPM, and EGPA, while exploring the common genetic, immunological, and pathological conditions among these adult-onset eosinophilic diseases.

Key words

Asthma; Eosinophils; Extracellular trap cell death; Thymic stromal lymphopoietin

1. Introduction

Allergic diseases in the upper and the lower airways such as allergic rhinitis and atopic asthma are usually accompanied by local eosinophilia and sometimes by peripheral blood eosinophilia. Chronic rhinosinusitis with nasal polyps (CRSwNP) and non-atopic asthma are also characterized by local and systemic eosinophilia, however, there are substantial differences in the age of onset and the presence of atopy. Atopic asthma and allergic rhinitis develop during childhood in most cases, and mediated by type 1 hypersensitivity reactions to allergen(s), as are other childhood-onset allergic diseases such as food allergies and eczema. Patients often develop these diseases consecutively as an allergic march (Figure 1), suggesting the presence of common genetic backgrounds and pathophysiology.

In contrast to childhood-onset disease, adult-onset asthma frequently presents as a non-atopic and eosinophilic condition,¹⁻⁶ although it shares similar clinical and physiological features. This phenotype of asthma is closely associated with other adult-onset eosinophilic airway diseases such as nonsteroidal anti-inflammatory drug (NSAID)-exacerbated airway disease (N-ERD) and CRSwNP/eosinophilic CRS, as well as an eosinophilic systemic disease, eosinophilic granulomatosis with polyangiitis (EGPA) (Figure 1). Allergic bronchopulmonary aspergillosis/mycosis (ABPA/ABPM) is another adult-onset eosinophilic airway disease, although it also has an allergic component with sensitization to fungal allergens. It is highly likely that there exist some common genetic, immunological, and pathological conditions among these adult-onset eosinophilic diseases.

For Japan, a society ageing at a globally unprecedented pace, a comprehensive and cross-sectional study of adult-onset allergic and immunological diseases is quite indispensable. Therefore, the Strategic Outlook toward 2030: Japan's Research for Allergy and Immunology, established by a multi-disciplinary committee comprising the Ministry of Health, Labour and Welfare of Japan, clinical and basic researchers, patients, and public groups, has proposed that the promotion of research and development for adult-onset eosinophilic airway diseases is as important as that of childhood-onset allergic diseases.⁷ In this review, we will discuss the pathophysiology, genetics, and clinical features of late-onset eosinophilic asthma, N-ERD, ABPA/ABPM, EGPA, and CRSwNP/eosinophilic CRS.

2. Perspective from eosinophil biology

Eosinophils are short-lived, bone-marrow-derived granulocytes. Eosinophil differentiation is largely dependent on IL-5, which is produced by Th2 cells and group 2 innate lymphoid cells

(ILC2s). Although type 1, 2, and 3 inflammation may overlap and balancing real-life (reviewed in ref. 8), type 2 inflammation is closely associated with eosinophilic inflammation. In response to type 2 cytokines, terminally differentiated eosinophils are mobilised from a niche in the bone marrow into the blood circulation. In a normal homeostatic condition, the half-life of blood eosinophils is approximately 25 h, and the majority of senescent (or apoptotic) eosinophils are cleared in the liver and spleen, where they are taken up by macrophages of the reticuloendothelial system.⁹ Apoptotic eosinophil clearance by phagocytotic cells (efferocytosis) mediates the resolution of inflammation and is shown to be dependent on the receptor tyrosine kinase Mer.¹⁰ Although the life span of eosinophils within the tissue is unknown, it is estimated to be less than 1 week in homeostatic conditions.¹¹ Tissue presence of eosinophils is determined based on recruitment, retention, and clearance (Figure 2). Eosinophil express various surface receptors and stored presynthesised specific granule proteins and mediators in their crystalloid granules, indicating that they can regulate immune system immediately within minutes of stimulus-induced cell activation (reviewed in ref. 11-13).

Tissue eosinophil migration through the epithelium into the lumen such as bronchi and sinuses, is a hallmark of pathological conditions. In inflamed tissue and lumen, eosinophils may undergo an active non-apoptotic cell death termed eosinophil ETosis (EETosis)¹⁴. EETosis is associated with plasma membrane rupture and release of free granules along with histone-retained chromatin structure, i.e. eosinophil extracellular traps (EETs)^{9,15,16}. EETs are considered an innate immune system to prevent dissemination of pathogens, but their excess formation could be pathogenic. EETs form stable aggregates that contribute to the viscosity of secretions observed in eosinophilic

55 CRS,¹⁵ eosinophilic otitis media,¹⁷ and ABPA.¹⁸ In a mouse model, EETs stimulated the production
56 of epithelial cytokines including thymic stromal lymphopoietin (TSLP) and IL-33.¹⁹

57 Localised autoimmune phenomenon was associated with disease severity or sub-optimal control
58 in asthma and EGPA.²⁰⁻²² Interestingly, sputum autoantibodies against eosinophil peroxidase,
59 identified often in severe asthmatics, can directly induce EETosis *in vitro*.²⁰ EETosis is also
60 associated with the crystallisation of galectin-10 to form Charcot-Leyden crystals (CLCs), a
61 classical hallmark of eosinophilic inflammation.²³⁻²⁵ CLCs are recognised by the NLRP3
62 inflammasome, which may sustain inflammation that follows eosinophilic inflammatory
63 processes.²⁶ Persson et al. advocated a novel concept of “CLC crystallopathy” by showing that
64 antibodies directed against key epitopes of the CLC crystallisation interface dissolved CLCs and
65 reversed crystal-driven inflammation, IgE synthesis, and bronchial hyperreactivity in a humanised
66 mouse model of asthma.²³

67 The presence of eosinophils with different properties is an attractive view in explaining the
68 diversity of eosinophilic diseases. The functional plasticity of recruited eosinophils is influenced by
69 the dynamic tissue microenvironment changes between homeostatic and disease states.²⁷

70 Lipidomic analysis for searching human eosinophils has shown that eosinophils from severe
71 asthma and CRSwNP represented a dysregulated fatty acid metabolic profile with decreased
72 pro-resolving protectin D1 and increased LTD₄ synthesis.^{28,29} In a mouse model, lung
73 tissue-resident eosinophils and eosinophils recruited upon type 2 inflammation are phenotypically
74 distinct populations.³⁰

75 Many eosinophil-derived cytokines are found as preformed mediators packaged within eosinophil
76 granules, suggesting that the cytokines may regulate the immune system immediately in their
77 microenvironment. Eosinophils can be a rich source of type 2 cytokines; two recent reports using
78 mice in different disease settings have demonstrated the pathological importance of
79 eosinophil-derived IL-4.^{31,32} Chronic tissue eosinophilia initiate tissue remodeling through their
80 secretory products, including VEGF, TGF β , and profibrotic osteopontin.^{11,33,34} In a cohort of
81 moderate persistent asthmatics, blood eosinophil count and local granule protein concentration
82 were closely accompanied with an increased airway sensory innervation, a key mechanism for
83 increased airway responsiveness.³⁵ Recent evidence indicates that excessive eosinophil
84 transepithelial migration leads to airway obstruction. The presence of luminal eosinophils in
85 severe asthmatics has been linked to mucus obstruction,³⁶ and eosinophil granule protein
86 generated oxidants may mediate mucus plug formation.³⁷

3. Genetic background of adult-onset eosinophilic airway diseases

Recent genome-wide association studies (GWASs) of adult-onset asthma, EGPA, CRSwNP, have convincingly identified a number of susceptibility loci (Supplemental Table 1).³⁸⁻⁴² The reported or mapped genes, which were associated with these asthma-related phenotypes in at least 2 GWASs, are listed in Table 1.

(a) GWAS of age of asthma onset

Recent large-scale GWASs of age of asthma onset revealed that adult-onset asthma is associated with fewer genes and their effect sizes at the loci are smaller than childhood-onset asthma.^{38,41,42} Pividori et al. found the first adult-onset asthma-specific association at 2q22.3 and identified a potential candidate gene, a long intergenic non-coding RNA.⁴¹ Enrichment studies showed genes at the adult-onset-specific loci were most highly expressed in the lung, blood, small intestine, and spleen.⁴¹ The findings suggest that adult-onset asthma is more lung-centred and is influenced by various environmental factors.^{41,43} Another GWAS identified a substantial positive correlation between body mass index and later-onset asthma, and Mendelian randomisation suggested that the body mass index causally increases the risk of asthma.⁴²

(b) GWAS of EGPA

A recent GWAS of EGPA reported a clinically and genetically distinct EGPA subset.⁴⁰ Myeloperoxidase-antineutrophil cytoplasmic antibody (ANCA)-positive EGPA, which revealed a genome-wide significant association at HLA-DQ, is an eosinophilic autoimmune disease, and ANCA-negative EGPA may have a mucosal/barrier dysfunction. Most susceptibility variants of EGPA are also associated with the eosinophil count in normal individuals, and Mendelian randomisation suggests that a primary tendency to eosinophilia contributes to the risk of EGPA. Several drug targets, *IL5*, *TSLP*, *BCL2*, and *CDK6*, were implicated by the GWAS, and further studies considering the two subtypes are necessary.

(c) GWAS of nasal polyps and CRS

A recent GWAS on nasal polyps (NP) and CRS found a total of 10 loci (*ALOX15*, *HLA*, *IL18R1-IL1RL1*, *IL33*, *FOXP1*, *CYP2S1*, *SLC22A4*, *MYRF*, *TSLP*, and 10p14) associated with NP at a genome-wide significance.³⁹ Two loci (*ALOX15*, *HLA*) corresponding to signals identified in the NP GWAS were also associated with CRS. A missense variant in *ALOX15* (p.Thr560Met), resulting in near total loss of 15-lipoxygenase activity, showed the largest protective effects against the risk of both NP and CRS. A recent study using single-cell RNA sequencing has shown that *ALOX15* was predominantly expressed by apical epithelial cells, and significantly increased in

124 NPs of N-ERD patients compared to NPs of CRSwNP patients.⁴⁴ The expression levels
125 significantly correlated with sinus disease severity and were associated with the presence of
126 asthma.⁴⁴ A downstream product of 15-lipoxygenase (15-oxo-EET) was elevated in NP compared
127 with that in healthy sinonasal tissue, and these findings suggest the involvement and interactions
128 of epithelial and mast cells in the transmetabolic process.⁴⁴ IL-13 further contributes to tissue
129 eosinophilia through eotaxin-3 production from fibroblasts and epithelial cells,⁴⁵ reportedly in a
130 15-lipoxygenase-1-dependent manner.⁴⁶

131

132 **(d) TSLP/IL33: possible common genetic factor for adult-onset eosinophilic airway** 133 **diseases**

134 TSLP is an essential epithelial cell-derived cytokine induced by allergens, pollutants, bacteria, and
135 viruses.⁴⁷ A recent study showed that double-stranded RNA-stimulated lung endothelial cells
136 produce both IL-33 and TSLP higher than those of bronchial epithelial cells.⁴⁸ In the GWAS on NP
137 and CRS, the top signal at the *TSLP* locus was rs1837253,³⁹ which is also associated with
138 ANCA-negative EGPA at genome-wide significant levels.⁴⁰ Rs1837253 (T/C) is located
139 immediately upstream of *TSLP*, and the frequencies of rs1837253 greatly differ across various
140 ethnic populations (<https://www.ncbi.nlm.nih.gov/variation/tools/1000genomes/>). GWASs,
141 including meta-analyses have also identified associations of rs1837253 (risk allele C) and asthma
142 in various ethnic groups, asthma-related phenotypes, and eosinophilia.⁴⁹⁻⁵² A recent study
143 reported a significant association of the rs1837253 genotype with CRSwNP and N-ERD, and
144 demonstrated a positive correlation between the risk allele and the number of eosinophils in
145 mucosal tissues of patients with CRSwNP in the Japanese population.⁵³

146 Recent GWASs have convincingly identified associations of the genetic variants at the *IL33* with
147 adult-onset asthma and nasal polyps.^{38,39,41,42} Genetic signals at the *IL33* predominantly
148 associated with blood eosinophil levels in the general population and with an eosinophilic asthma
149 phenotype and also influenced *IL33* levels in the airway epithelium, with the risk allele associating

with increased *IL33 in vivo*.⁵⁴ An epigenetic analysis of human naïve ILC2s activated with IL-25 and IL-33 in vitro revealed that a number of asthma-associated genetic variants localised to ILC2-specific regulatory elements.⁵⁵

153

4. Clinical characteristics and pathophysiology of adult-onset eosinophilic airway diseases

(a) Adult-onset, eosinophil-predominant phenotype of severe asthma

The adult-onset eosinophil-predominant severe asthma phenotype was first demonstrated in a UK study as the “late-onset inflammation predominant” phenotype with a high proportion of eosinophils in the sputum.¹ A severe asthma research program in the United States identified a late-onset, non-atopic phenotype in obese women,⁵⁶ and the following studies demonstrated that CRSwNP and impaired pulmonary functions are major comorbidities in this phenotype.^{5,6} A similar late-onset phenotype with peripheral blood eosinophilia, CRSwNP, and poor pulmonary functions was identified in the European (U-BIOPRED), Chinese, and Japanese cohorts.²⁻⁴ There is some inconsistency with gender dominance; a Japanese report suggests that there are two sub-phenotypes: a male-dominant sub-phenotype with cigarette smoking and an obese female-dominant sub-phenotype.²

This late-onset eosinophilic phenotype accounts for 20-40% of severe asthma cases (Table 2).¹⁻⁶ Peripheral blood eosinophilia >300/μL is observed more commonly (40-50%); however, eosinophilia is associated with poor asthma control and frequent exacerbation only in adult-onset severe asthmatics, but not in patients with mild to moderate disease⁵⁷ or in children.⁵⁸ Interestingly, peripheral blood eosinophil counts were associated with type 2 gene expression in sputum cells only in the non-obese patients age 35 years or older.⁵⁹ A Japanese study also confirmed that sputum IL-5 concentrations were increased only in adult-onset phenotypes but not in childhood-onset eosinophilic asthma.²

Omics approaches have been uncovering the pathophysiology of adult-onset eosinophil-predominant asthma. A gene expression cluster analysis using sputum cells revealed that a population with enhanced expression of the IL-33 receptor gene (*IL1RL1*) and TSLP receptor gene (*CRLF*) exhibited a phenotype consistent with adult-onset eosinophil-predominant asthma.⁶⁰ Other researchers confirmed an increased expression of *IL1RL1*⁶¹ or *CRLF*⁶² in the patients with this phenotype. Transcriptome analysis of bronchial epithelial cells collected by bronchoscopy has demonstrated that the IL-13 signature is associated with a phenotype with peripheral blood eosinophilia as expected,^{63,64} but the IL-6 signature, which impairs epithelial

182 barrier function and induces IL-33 expression, is also correlated with peripheral blood eosinophilia
183 and frequent asthma exacerbation.⁶⁵
184 Peripheral blood eosinophilia is the most important biomarker for predicting the efficacy of
185 anti-IL-5 antibodies (mepolizumab and reslizumab) and an anti-IL-5 receptor- α antibody
186 (benralizumab).⁶⁶⁻⁶⁸ Later age of asthma onset, CRSwNP, and low pulmonary functions, the
187 features compatible with adult-onset eosinophilic asthma phenotype, correlate well with better
188 responses to benralizumab treatment.⁵⁹ However, the fraction of exhaled nitric oxide (FeNO),
189 another type 2 biomarker, does not correlate with the effect of mepolizumab; FeNO-high,
190 eosinophil-low patients did not respond to mepolizumab treatment.⁶⁷ Peripheral blood eosinophilia
191 is also a biomarker of efficacy for an anti-IL-4 receptor- α antibody (dupilumab).^{69,70} Regarding an
192 anti-IgE antibody (omalizumab), a recent study showed no relationship between peripheral blood
193 eosinophil counts and its clinical usefulness.⁷¹

194

195 **(b) NSAID-exacerbated respiratory disease**

196 N-ERD is an NSAID hypersensitivity that causes rapid airway obstruction.⁷² The natural history of
197 N-ERD begins at the mean age of 35 years, with NPs (especially hyposmia) and bronchial
198 symptoms, followed by NSAIDs hypersensitivity within a few years. Serum total IgE levels are
199 increased, but atopic constitution is usually absent (Table 2).

200 Cysteinyl leukotriene (cysLT) and prostaglandin (PG) D₂ overproduction, especially in NPs, is
201 associated with the pathological condition.⁷³ Chronic reduction of PGE₂ synthesis due to
202 cyclooxygenase (COX)-2 and EP2 receptor dysfunction leads to the overproduction of cysLT,^{74,75}
203 reduction of anti-inflammatory mediators such as lipoxin,⁷⁶ and activation of mast cells and
204 ILC2s.^{74,77,78} Pre-inhalation of PGE₂⁷⁹ or cigarette smoking⁸⁰ suppresses N-ERD, supporting that
205 COX-2 hypofunction and PGE₂ loss are essential pathogenesis of N-ERD. Enhanced production
206 of IL-33, TSLP, and IL-25 further activates mast cells and ILC2s.⁸¹⁻⁸³

207 Peripheral blood eosinophilia is commonly observed in patients with N-ERD.⁸⁴ During aspirin
208 provocation, the number of blood eosinophils decreases,⁸⁵ and activated eosinophils accumulate
209 in the respiratory tract,⁸⁶ although the biomarkers of eosinophil activation such as 3-bromotyrosine
210 and eosinophil-derived neurotoxin in urine do not increase.⁸⁷ Another unique inflammatory
211 condition in N-ERD is the presence of eosinophils or other granulocytes in peripheral blood and
212 airway tissues adhered to platelets,^{88,89} which provide LTA4 to granulocytes and participate in
213 CysLT overproduction.⁷⁴ A clinical trial with an antiplatelet drug, P2Y₁₂ receptor antagonist, failed
214 to demonstrate significant clinical improvement⁹⁰; however, future trials are expected for other
215 drugs targeting P-selectin, whose expression is particularly enhanced in platelets of N-ERD
216 patients.⁸⁹

217 The most characteristic pathological condition of N-ERD is prominent mast cell inflammation. Mast
218 cell activation biomarkers are increased during aspirin challenge, significantly correlating with
219 decreased lung function and increased urinary cysLT concentration.^{73,91,92} Airway reactivity to
220 adenosine monophosphate, a specific mast cell stimulus, is increased,⁹³ whereas cromolyn, a
221 mast cell stabiliser, shows specific bronchodilation.⁹⁴

222 The role of mast cells is further confirmed by a randomised controlled trial, which has
223 demonstrated that omalizumab reduces mast cell activation, aspirin sensitivity, and cysLT
224 overproduction.⁹⁵ Mepolizumab shows improvement in nasal symptoms and asthma control in
225 patients with N-ERD.⁹⁶ Dupilumab has been reported to be effective for upper respiratory
226 symptoms of N-ERD.⁹⁷ Future progress is also expected for TSLP and IL-33 antagonists.⁹⁴

227

228

229 (c) Allergic bronchopulmonary aspergillosis/mycosis

230 ABPA/ABPM develops via type I and III hypersensitivity reactions to filamentous fungi.⁹⁸ Fungal
231 colonisation in the intrabronchial mucus is essential for the development of type III
232 hypersensitivity. *A. fumigatus* with its small conidia size and thermotolerance is the dominant
233 causal fungi, although fungi with similar microbiological features such as *Penicillium* spp. and
234 *Shizophyllum commune* can also cause ABPM.^{99,100} Asthma-related ABPM rarely occurs during
235 childhood (Table 2), although a substantial proportion of children with asthma are sensitised to *A.*
236 *fumigatus* and other fungi.¹⁰¹ Actually, two-thirds of patients with ABPM in a Japanese nation-wide
237 survey developed the disease at the age of 50 years or older.¹⁰² Adult asthmatics exposed to
238 higher dose of inhaled corticosteroids are susceptible to bronchial colonisation of *A. fumigatus*,¹⁰³
239 and *de novo* sensitisation to *A. fumigatus* can occur in adults with established asthma, especially
240 in those with severe disease treated with high-dose inhaled corticosteroids and poor pulmonary
241 functions.¹⁰⁴

242 Robust type I hypersensitivity reaction and high IgE levels in serum are characteristics of ABPA,
243 but not necessarily of non-*Aspergillus* ABPM.¹⁰⁵ A universal and cardinal histological feature of
244 ABPA/ABPM is the presence of allergic mucin enriched with eosinophils harbouring fungal hyphae
245 in the central bronchi.^{106,107} Allergic mucin in ABPM is associated with EETosis as discussed
246 previously in section 2,¹⁸ and this highly viscous mucin stretches the inflamed airway wall and
247 spreads fungal components transluminally, causing peripheral lung lesions. Allergic mucin
248 containing fungal hyphae, which presents with “peanut butter-like” viscosity and high attenuation
249 on CT, is also observed in paranasal sinus of the patients with allergic fungal rhinosinusitis,
250 suggesting a similar pathophysiology between these two conditions.¹⁰⁸

Systemic corticosteroids and/or anti-fungal drugs are the standard treatment for ABPM; however, flares and recurrences of the disease are quite common, and alternative treatments are warranted to avoid adverse effects and emergence of drug-resistant fungi. Omalizumab has been demonstrated to suppress exacerbation rate and reduce the dose of systemic corticosteroids, but radiographic improvement is limited.¹⁰⁹⁻¹¹² In contrast, mepolizumab or benralizumab effectively control symptoms and clear the mucus plugs in the bronchi.^{113,114} These observations support the hypothesis that the major pathophysiology of ABPM is the formation of eosinophilic mucus plugs in the airways. There are a few case reports showing that dupilumab improves residual asthmatic symptoms after mepolizumab treatment.¹¹⁵

260

261 **(d) Eosinophilic chronic rhinosinusitis**

The inflammatory pattern of CRSwNP is diverse geographically and ethnically.¹¹⁶ While the pathological features of CRSwNP show type 2 inflammation and eosinophil infiltration in Western countries, Japanese CRSwNP has shown type 1 and type 3 inflammation and neutrophil infiltration as recently as 25 years ago. Since the late 1990s, the incidence of CRSwNP with strong eosinophil infiltration has increased in Japan.¹¹⁷ Strong eosinophil-infiltrated NP recurs soon after surgery and is difficult to treat. This intractable/recalcitrant CRSwNP has been termed eosinophilic CRS, and therefore, eosinophilic CRS without NP has not been included in this category.¹¹⁷

The Japanese Epidemiological Survey of Refractory Eosinophilic Chronic Rhinosinusitis (JESREC) study established diagnosis criteria and severity classifications for eosinophilic CRS (Figure E1).¹¹⁸ A definite diagnosis of eosinophilic CRS can be made by tissue eosinophilia (eosinophil count ≥ 70 /HPF). Blood eosinophils, ethmoid disease, asthma, and N-ERD were all associated with severity of ECRS. Both mild and moderate eosinophilic CRS are similar to the moderate type 2 group of CRSwNP, whereas severe eosinophilic CRS seems to be the severe type 2 group of CRSwNP in Western countries. The clinical features of eosinophilic CRS include bilateral NPs, loss of smell, nasal obstruction, viscous rhinorrhea, and high comorbidities of asthma (24-67%) and N-ERD (7-16%) (Table 2).^{118,119}

The pathological feature of NPs is a fibrin deposition accompanied by the dysregulation of coagulation and fibrinolysis cascade.^{120,121} Eosinophils in NPs release tissue factor (TF), which is an initiator of extrinsic coagulation cascade.^{122,123} The levels of thrombin and factor XIIIa are elevated in NPs and act in the final step of the formation of fibrin mesh.^{124,125} Tissue plasminogen activator (tPA) is downregulated in NPs¹²¹ by the hypermethylation of the proximal tPA gene promoter in NPs.¹²⁶ Thrombin-activatable fibrinolysis inhibitor (TAFI) inhibits the fibrinolysis cascade. The levels of TAFI are elevated in CRSwNP, especially CRSwNP with asthma.¹²⁵ These results suggest that targeting for fibrin is a promising strategy for treating NPs.¹²⁷

286 To find biomarkers and new pathological mechanisms of eosinophilic CRS, RNA sequencing
287 was performed to identify differentially expressed genes between NP and inferior turbinate.¹²⁸ The
288 expression of *POSTN*, *CST-1*, *CCL18*, *CXCL17*, and *ALOX15* was significantly increased in NP.
289 *CST-1*, a cystatin protease inhibitor, is produced by nasal epithelial cells (Figure 2). *CST-1*
290 showed greater expression in eosinophilic CRS than non-eosinophilic CRS. The expression of
291 *CST-1* mRNA in NPs displayed a significant positive correlation with the eosinophil count in NP
292 and with expression level of mRNAs for TSLP, IL-33, and eotaxin-1. *CST-1* enhanced production
293 of TSLP and eotaxin-1 by nasal compartment cells, suggesting that *CST-1* is involved in the
294 promotion of eosinophilic infiltration in patients with eosinophilic CRS.¹²⁹

295 In general, oral corticosteroids are the only effective conservative therapy. Type 2 targeting
296 biologics are emerging treatment options for severely uncontrolled patients with eosinophilic
297 CRS.^{130,131} Dupilumab,^{130,132} mepolizumab,^{9,133} benralizumab,¹³⁴ and omalizumab¹³⁵ are effective
298 treatments for eosinophilic CRS. Dupilumab reduced NP, sinus opacification, and symptom
299 severity in a phase III clinical study. Similar effects of dupilumab were observed in Japanese
300 patients with CRSwNP, indicating no differences by ethnicity/race in efficacy of biologics.
301 Benralizumab completely suppresses blood eosinophil counts in all cases, but nasal polyp scores
302 in 50% of eosinophilic CRS were not reduced. This result suggested that eosinophils are not the
303 only pathogenesis for NP of eosinophilic CRS.

304

305 **(e) Eosinophilic granulomatosis with polyangiitis**

306 EGPA is a rare disease characterised by severe eosinophilia, asthma, and systemic vasculitis and
307 most often occurs between the ages of 30 and 70 years (mean 50 years) (Table 2). Severe
308 eosinophilic airway inflammation precedes developing vasculitis by several years.¹³⁶⁻¹³⁹ The
309 characteristics of the respiratory symptoms are basically non-atopic eosinophilic asthma, which is
310 associated with eosinophilic CRS and partly with eosinophilic pneumonia/bronchiolitis.^{136,140} When
311 vasculitis develops, severe eosinophilic inflammation and ischaemic damage coexist in systemic
312 organs. The positive rate of ANCA is only 30-35%. Glomerulonephritis is more likely to occur in the
313 ANCA-positive group, and cardiac dysfunction is more common in the negative group,^{40,141} but
314 there is no clear difference in respiratory symptoms and eosinophilia between them.^{136,140}

315 In EGPA patients, serum total IgE is often high, but allergen sensitisation is low.^{136,140} Asthma is
316 intractable and is associated with shortness of breath and cough but may not include wheezing.
317 Unlike usual asthma, the airway pathophysiology of EGPA may involve more peripherally
318 predominant inflammation and persistent airflow limitation.^{136,140}

319 In EGPA, three different pathological conditions, eosinophilic inflammation, vasculitis, and
320 granuloma formation, exist simultaneously. GM-CSF; IL-3, -4, -5, -13, -25, and -33; and TSLP are

the cytokines that induce type 2 inflammation of EGPA,^{137-139,142,143} and an enhanced release of B-cell activation factor BAFF causes IgE and IgG4 production (Figure 2).^{137,139,143,144} In addition to type 2 inflammation, enhanced production of IL-17 and INF- γ from Th17 and Th1 cells decreases in regulatory T cells and increases in ILC2s coexists, complicating the airway inflammation of EGPA.^{142,145} IL-5 and eotaxin-3 levels are significantly increased in bronchoalveolar lavage fluid (BALF) in patients with acute phase EGPA, and IL-5, IL-4, and IL-10 gene expression were enhanced in BALF cells.¹⁴⁶ Furthermore, a large amount of IL-5 is produced by cultured peripheral blood mononuclear cells of stable EGPA patients.¹⁴⁷ ANCA activates neutrophils, which attack the vascular wall via degranulation and the formation of NETs.¹³⁷ However, the role of ANCA and interaction between ANCA and type 2 inflammation remain unclear. Biomarkers for EGPA include sIL2R, eotaxin-3, GM-CSF, IgG4, 12-HETE, and urinary LTE4 (Table 2).^{138,139,143,145,148-150} The standard treatment for EGPA is a combination of systemic corticosteroids and immunosuppressants,¹³⁸ but nevertheless, there are many cases of persistent vascular and respiratory symptoms. Mepolizumab suppresses not only eosinophilic inflammation but also relapse of vascular symptoms.¹⁵¹ The MIRRA study reported that administration of mepolizumab for 1 year could suppress relapse of vasculitis and reduce corticosteroid dosage to 40%.¹⁵¹ A similar international clinical trial for benralizumab has started. Omalizumab is effective in the respiratory symptoms of EGPA, but its effect on vascular inflammation is unclear.¹⁵² A high response rate for EGPA in ANCA-positive cases with rituximab was reported in an observational study,⁴⁰ and an international clinical trial is underway.

5. Conclusion

We propose a concept of “adult-onset eosinophilic airway diseases” including asthma, N-ERD, ABPA/ABPM, CRSwNP/eosinophilic CRS, and EGPA. Common pathophysiology such as EETosis/mucus plug formation and common genetic backgrounds including TSLP/IL-33 pathway genes seem to underlie these diseases^{153,154}. In addition, each disease exhibits a specific pathophysiology such as mast cell and platelet activation in N-ERD, fungal sensitisation in ABPA/ABPM, abnormality in coagulation/fibrinolysis pathway in CRSwNP/eosinophilic CRS, and autoimmunity in EGPA. The clinical efficacy of anti-IL-5 treatments was also variable among the diseases; high for eosinophilic asthma, ABPA/ABPM, and EGPA, whereas low for N-ERD and CRSwNP/eosinophilic CRS (Table 1). We still do not know how these diseases develop only in adulthood, and further studies such as deep phenotyping and integration of multi-omics data of this unique disease complex are warranted in the era of precision medicine.

358 **Conflicts of interest**

359 Dr. Asano reports grants from Japan Agency for Medical Research and Development, during the
360 conduct of the study; personal fees from Teijin Pharma Ltd., personal fees from Novartis Pharma
361 K.K., personal fees from Sanofi S. A., personal fees from Thermo Fisher Diagnostics K.K.,
362 personal fees from AstraZeneca K.K., personal fees from GlaxoSmithKline K.K., personal fees
363 from Nippon Boehringer Ingelheim Co., outside the submitted work; Dr. Ueki reports grants from
364 Mochida Memorial Foundation, grants from JSPS KAKENHI, grants and personal fees from
365 AstraZeneca K.K, grants and personal fees from Novartis, grants from Maruho Co. Ltd., grants
366 and personal fees from GlaxoSmithKline K.K., during the conduct of the study; Dr. Tamari
367 received lecture fees from AstraZeneca K.K., Kyorin Pharmaceutical Co., Ltd., and Eisai Co., Ltd.;
368 Dr. Imoto has nothing to disclose; Dr. Fujieda reports grants from Maruho Co.,Ltd., grants from
369 Tsumura & Co., grants and personal fees from Mitsubishi Tanabe Pharma Corporation, grants and
370 personal fees from Sanofi S.A., personal fees from KYORIN Pharmaceutical Co., Ltd., personal
371 fees from TAIHO Pharmaceutical Co., Ltd., personal fees from GlaxoSmithKline K.K., personal
372 fees from Kyowa Kirin Co., Ltd. outside the submitted work; Dr. Taniguchi reports personal fees
373 from GlaxoSmithKline K.K., personal fees from AstraZeneca K.K., personal fees from Sanofi S. A.,
374 outside the submitted work.

375

References

1. Haldar P, Pavord ID, Shaw DE, et al. Cluster analysis and clinical asthma phenotypes. *Am J Respir Crit Care Med* 2008;178:218-224.
2. Konno S, Taniguchi N, Makita H, et al. Distinct phenotypes of smokers with fixed airflow limitation identified by cluster analysis of severe asthma. *Ann Am Thorac Soc* 2018;15:33-41.
3. Lefaudeux D, De Meulder B, Loza MJ, et al. U-BIOPRED clinical adult asthma clusters linked to a subset of sputum omics. *J Allergy Clin Immunol* 2017;139:1797-1807.
4. Qiu R, Xie J, Chung KF, et al. Asthma phenotypes defined from parameters obtained during recovery from a hospital-treated exacerbation. *J Allergy Clin Immunol Pract* 2018;6:1960-1967.
5. Wu W, Bang S, Bleecker ER, et al. Multiview cluster analysis identifies variable corticosteroid response phenotypes in severe asthma. *Am J Respir Crit Care Med* 2019;199:1358-1367.
6. Wu W, Bleecker E, Moore W, et al. Unsupervised phenotyping of Severe Asthma Research Program participants using expanded lung data. *J Allergy Clin Immunol* 2014;133:1280-1288.
7. Adachi T, Kainuma K, Asano K, et al. Strategic outlook toward 2030: Japan's research for allergy and immunology - Secondary publication. [published ahead of print June 26, 2020]. *Allergol Int* doi: 10.1016/j.alit.
8. Sonnenberg GF, Hepworth MR. Functional interactions between innate lymphoid cells and adaptive immunity. *Nat Rev Immunol*. 2019;19:599-613.
9. Nagase H, Ueki S, Fujieda S. The roles of IL-5 and anti-IL-5 treatment in eosinophilic diseases: Asthma, eosinophilic granulomatosis with polyangiitis, and eosinophilic chronic rhinosinusitis. *Allergol Int* 2020;69:178-186.
10. Felton JM, Lucas CD, Dorward DA, et al. Mer-mediated eosinophil efferocytosis regulates resolution of allergic airway inflammation. *J Allergy Clin Immunol* 2018;142:1884-1893 e1886.
11. Klion AD, Ackerman SJ, Bochner BS. Contributions of eosinophils to human health and disease. *Annu Rev Pathol* 2020;15:179-209.
12. Melo RCN, Weller PF. Contemporary understanding of the secretory granules in human eosinophils. *J Leukoc Biol*. 2018;104:85-93.

- Accepted Article
13. Weller PF, Spencer LA. Functions of tissue-resident eosinophils. *Nat Rev Immunol*. 2017;17:746-760.
 14. Ueki S, Melo RC, Ghiran I, et al. Eosinophil extracellular DNA trap cell death mediates lytic release of free secretion-competent eosinophil granules in humans. *Blood*. 2013;121:2074-2083.
 15. Ueki S, Konno Y, Takeda M, et al. Eosinophil extracellular trap cell death-derived DNA traps: Their presence in secretions and functional attributes. *J Allergy Clin Immunol* 2016;137:258-267.
 16. Kim HJ, Sim MS, Lee DH, et al. Lysophosphatidylserine induces eosinophil extracellular trap formation and degranulation: Implications in severe asthma. *Allergy*. 2020.
 17. Ohta N, Ueki S, Konno Y, et al. ETosis-derived DNA trap production in middle ear effusion is a common feature of eosinophilic otitis media. *Allergol Int* 2018;67:414-416.
 18. Muniz VS, Silva JC, Braga YAV, et al. Eosinophils release extracellular DNA traps in response to *Aspergillus fumigatus*. *J Allergy Clin Immunol* 2018;141:571-585 e577.
 19. Choi Y, Kim YM, Lee HR, et al. Eosinophil extracellular traps activate type 2 innate lymphoid cells through stimulating airway epithelium in severe asthma. *Allergy* 2020;75:95-103.
 20. Mukherjee M, Bulir DC, Radford K, et al. Sputum autoantibodies in patients with severe eosinophilic asthma. *J Allergy Clin Immunol* 2018;141:1269-1279.
 21. Mukherjee M, Forero DF, Tran S, et al. Sub-optimal treatment response to anti-IL-5 monoclonal antibodies in severe eosinophilic asthmatics with airway autoimmune phenomena. [published ahead of print May 22, 2020] *Eur Respir J* doi: 10.1183/13993003.00117-2020.
 22. Qin R, Long F, Xiao X, et al. Sputum autoantibodies are more relevant in autoimmune responses in asthma than are serum autoantibodies. *Allergy Asthma Immunol Res* 2019;11:406-421.
 23. Persson EK, Verstraete K, Heyndrickx I, et al. Protein crystallization promotes type 2 immunity and is reversible by antibody treatment. *Science* 2019;364.
 24. Ueki S, Tokunaga T, Melo RCN, et al. Charcot-Leyden crystal formation is closely associated with eosinophil extracellular trap cell death. *Blood* 2018;132:2183-2187.

25. Melo RCN, Wang H, Silva TP, et al. Galectin-10, the protein that forms Charcot-Leyden crystals, is not stored in granules but resides in the peripheral cytoplasm of human eosinophils. *J Leukoc Biol*. 2020;108:139-149.
26. Rodriguez-Alcazar JF, Ataide MA, Engels G, et al. Charcot-Leyden crystals activate the NLRP3 inflammasome and cause IL-1 β inflammation in human macrophages. *J Immunol* 2019;202:550-558.
27. Abdala-Valencia H, Coden ME, Chiarella SE, et al. Shaping eosinophil identity in the tissue contexts of development, homeostasis, and disease. *J Leukoc Biol* 2018;104:95-108.
28. Miyata J, Fukunaga K, Kawashima Y, et al. Dysregulated fatty acid metabolism in nasal polyp-derived eosinophils from patients with chronic rhinosinusitis. *Allergy* 2019;74:1113-1124.
29. Miyata J, Fukunaga K, Iwamoto R, et al. Dysregulated synthesis of protectin D1 in eosinophils from patients with severe asthma. *J Allergy Clin Immunol* 2013;131:353-360 e351-352.
30. Mesnil C, Raulier S, Paulissen G, et al. Lung-resident eosinophils represent a distinct regulatory eosinophil subset. *J Clin Invest* 2016;126:3279-3295.
31. Lee SH, Chaves MM, Kamenyeva O, et al. M2-like, dermal macrophages are maintained via IL-4/CCL24-mediated cooperative interaction with eosinophils in cutaneous leishmaniasis. *Sci Immunol* 2020;5:eaaz4415.
32. Diny NL, Baldeviano GC, Talor MV, et al. Eosinophil-derived IL-4 drives progression of myocarditis to inflammatory dilated cardiomyopathy. *J Exp Med* 2017;214:943-957.
33. Takemura N, Kurashima Y, Mori Y, et al. Eosinophil depletion suppresses radiation-induced small intestinal fibrosis. *Sci Transl Med* 2018;10.
34. Morimoto Y, Hirahara K, Kiuchi M, et al. Amphiregulin-producing pathogenic memory T helper 2 cells instruct eosinophils to secrete osteopontin and facilitate airway fibrosis. *Immunity* 2018;49:134-150 e136.
35. Drake MG, Scott GD, Blum ED, et al. Eosinophils increase airway sensory nerve density in mice and in human asthma. *Sci Transl Med* 2018;10.
36. Svenningsen S, Eddy RL, Lim HF, Cox PG, Nair P, Parraga G. Sputum eosinophilia and magnetic resonance imaging ventilation heterogeneity in severe asthma. *Am J Respir Crit Care Med* 2018;197:876-884.
37. Dunican EM, Elicker BM, Gierada DS, et al. Mucus plugs in patients with asthma linked to eosinophilia and airflow obstruction. *J Clin Invest* 2018;128:997-1009.
38. Ferreira MAR, Mathur R, Vonk JM, et al. Genetic architectures of childhood- and adult-onset asthma are partly distinct. *Am J Hum Genet* 2019;104:665-684.

39. Kristjansson RP, Benonisdottir S, Davidsson OB, et al. A loss-of-function variant in ALOX15 protects against nasal polyps and chronic rhinosinusitis. *Nat Genet* 2019;51:267-276.
40. Lyons PA, Peters JE, Alberici F, et al. Genome-wide association study of eosinophilic granulomatosis with polyangiitis reveals genomic loci stratified by ANCA status. *Nat Commun* 2019;10:5120.
41. Pividori M, Schoettler N, Nicolae DL, Ober C, Im HK. Shared and distinct genetic risk factors for childhood-onset and adult-onset asthma: genome-wide and transcriptome-wide studies. *Lancet Respir Med* 2019;7:509-522.
42. Zhu Z, Guo Y, Shi H, et al. Shared genetic and experimental links between obesity-related traits and asthma subtypes in UK Biobank. *J Allergy Clin Immunol* 2020;145:537-549.
43. Schoettler N, Rodriguez E, Weidinger S, Ober C. Advances in asthma and allergic disease genetics: Is bigger always better? *J Allergy Clin Immunol* 2019;144:1495-1506.
44. Stevens WW, Staudacher AG, Hulse KE, et al. Activation of the 15-lipoxygenase pathway in aspirin-exacerbated respiratory disease. [published ahead of print May 1, 2020] *J Allergy Clin Immunol* doi: 10.1016/j.jaci.2020.04.031.
45. Yamada T, Miyabe Y, Ueki S, et al. Eotaxin-3 as a plasma biomarker for mucosal eosinophil infiltration in chronic rhinosinusitis. *Front Immunol* 2019;10:74.
46. Li Z, Zeng M, Deng Y, et al. 15-Lipoxygenase 1 in nasal polyps promotes CCL26/eotaxin 3 expression through extracellular signal-regulated kinase activation. *J Allergy Clin Immunol* 2019;144:1228-1241 e1229.
47. Akdis CA, Arkwright PD, Bruggen MC, et al. Type 2 immunity in the skin and lungs. *Allergy* 2020;75:1582-1605.
48. Emi-Sugie M, Shoda T, Futamura K, et al. Robust production of IL-33 and TSLP by lung endothelial cells in response to low-dose dsRNA stimulation. [published ahead of print Apr 20, 2020] *J Allergy Clin Immunol* doi: 10.1016/j.jaci.2020.03.042.
49. Ferreira MA, Vonk JM, Baurecht H, et al. Shared genetic origin of asthma, hay fever and eczema elucidates allergic disease biology. *Nat Genet* 2017;49:1752-1757.
50. Astle WJ, Elding H, Jiang T, et al. The allelic landscape of human blood cell trait variation and links to common complex disease. *Cell* 2016;167:1415-1429 e1419.
51. Hirota T, Takahashi A, Kubo M, et al. Genome-wide association study identifies eight new susceptibility loci for atopic dermatitis in the Japanese population. *Nat Genet* 2012;44:1222-1226.

52. Torgerson DG, Ampleford EJ, Chiu GY, et al. Meta-analysis of genome-wide association studies of asthma in ethnically diverse North American populations. *Nat Genet* 2011;43:887-892.
53. Nakayama T, Hirota T, Asaka D, et al. A genetic variant near TSLP is associated with chronic rhinosinusitis with nasal polyps and aspirin-exacerbated respiratory disease in Japanese populations. *Allergol Int* 2020;69:138-140.
54. Ketelaar ME, Portelli MA, Dijk FN, et al. Phenotypic and functional translation of IL33 genetics in asthma. [published ahead of print May 19, 2020] *J Allergy Clin Immunol* doi: 10.1016/j.jaci.2020.04.051.
55. Stadhouders R, Li BWS, de Bruijn MJW, et al. Epigenome analysis links gene regulatory elements in group 2 innate lymphocytes to asthma susceptibility. *J Allergy Clin Immunol* 2018;142:1793-1807.
56. Moore WC, Meyers DA, Wenzel SE, et al. Identification of asthma phenotypes using cluster analysis in the Severe Asthma Research Program. *Am J Respir Crit Care Med* 2010;181:315-323.
57. Semprini R, Williams M, Semprini A, et al. Type 2 biomarkers and prediction of future exacerbations and lung function decline in adult asthma. *J Allergy Clin Immunol Pract* 2018;6:1982-1988 e1981.
58. Teague WG, Phillips BR, Fahy JV, et al. Baseline features of the severe asthma research program (SARP III) cohort: Differences with age. *J Allergy Clin Immunol Pract* 2018;6:545-554 e544.
59. Peters MC, Kerr S, Dunican EM, et al. Refractory airway type 2 inflammation in a large subgroup of asthmatic patients treated with inhaled corticosteroids. *J Allergy Clin Immunol* 2019;143:104-113 e114.
60. Kuo CS, Pavlidis S, Loza M, et al. T-helper cell type 2 (Th2) and non-Th2 molecular phenotypes of asthma using sputum transcriptomics in U-BIOPRED. *Eur Respir J* 2017;49:1602135.
61. Rossios C, Pavlidis S, Hoda U, et al. Sputum transcriptomics reveal upregulation of IL-1 receptor family members in patients with severe asthma. *J Allergy Clin Immunol* 2018;141:560-570.
62. Peters MC, Ringel L, Dyjack N, et al. A transcriptomic method to determine airway immune dysfunction in T2-High and T2-low asthma. *Am J Respir Crit Care Med* 2019;199:465-477.
63. Kuo CS, Pavlidis S, Loza M, et al. A transcriptome-driven analysis of epithelial brushings and bronchial biopsies to define asthma phenotypes in U-BIOPRED. *Am J Respir Crit Care Med* 2017;195:443-455.

64. Ostling J, van Geest M, Schofield JPR, et al. IL-17-high asthma with features of a psoriasis immunophenotype. *J Allergy Clin Immunol* 2019;144:1198-1213.
65. Jevnikar Z, Ostling J, Ax E, et al. Epithelial IL-6 trans-signaling defines a new asthma phenotype with increased airway inflammation. *J Allergy Clin Immunol* 2019;143:577-590.
66. FitzGerald JM, Bleecker ER, Menzies-Gow A, et al. Predictors of enhanced response with benralizumab for patients with severe asthma: pooled analysis of the SIROCCO and CALIMA studies. *Lancet Respir Med* 2018;6:51-64.
67. Shrimanker R, Keene O, Hynes G, Wenzel S, Yancey S, Pavord ID. Prognostic and predictive value of blood eosinophil count, fractional exhaled nitric oxide, and their combination in severe asthma: a post hoc analysis. *Am J Respir Crit Care Med* 2019;200:1308-1312.
68. Poznanski SM, Mukherjee M, Zhao N, et al. Asthma exacerbations on benralizumab are largely non-eosinophilic. [published ahead of print Jul 23, 2020] *Allergy*. doi: 10.1111/all.14514.
69. Casale TB, Chipps BE, Rosen K, et al. Response to omalizumab using patient enrichment criteria from trials of novel biologics in asthma. *Allergy* 2018;73:490-497.
70. Castro M, Corren J, Pavord ID, et al. Dupilumab efficacy and safety in moderate-to-severe uncontrolled asthma. *N Engl J Med* 2018;378:2486-2496.
71. Humbert M, Taille C, Mala L, et al. Omalizumab effectiveness in patients with severe allergic asthma according to blood eosinophil count: the STELLAIR study. *Eur Respir J* 2018;51:1702523.
72. Kowalski ML, Agache I, Bavbek S, et al. Diagnosis and management of NSAID-Exacerbated Respiratory Disease (N-ERD)-a EAACI position paper. *Allergy* 2019;74:28-39.
73. Higashi N, Taniguchi M, Mita H, Yamaguchi H, Ono E, Akiyama K. Aspirin-intolerant asthma (AIA) assessment using the urinary biomarkers, leukotriene E4 (LTE4) and prostaglandin D₂ (PGD₂) metabolites. *Allergol Int* 2012;61:393-403.
74. Boyce JA. Aspirin sensitivity: Lessons in the regulation (and dysregulation) of mast cell function. *J Allergy Clin Immunol* 2019;144:875-881.
75. Celejewska-Wojcik N, Wojcik K, Ignacak-Popiel M, et al. Subphenotypes of nonsteroidal antiinflammatory disease-exacerbated respiratory disease identified by latent class analysis. *Allergy*. 2020;75:831-840.
76. Perez-Novoa CA, Watelet JB, Claeys C, Van Cauwenberge P, Bachert C. Prostaglandin, leukotriene, and lipoxin balance in chronic rhinosinusitis with and without nasal polyposis. *J Allergy Clin Immunol* 2005;115:1189-1196.

77. Laidlaw TM, Boyce JA. Aspirin-exacerbated respiratory disease--new prime suspects. *N Engl J Med* 2016;374:484-488.
78. Doherty TA, Broide DH. Airway innate lymphoid cells in the induction and regulation of allergy. *Allergol Int* 2019;68:9-16.
79. Szczeklik A, Mastalerz L, Nizankowska E, Cmiel A. Protective and bronchodilator effects of prostaglandin E and salbutamol in aspirin-induced asthma. *Am J Respir Crit Care Med* 1996;153:567-571.
80. Hayashi H, Fukutomi Y, Mitsui C, et al. Smoking cessation as a possible risk factor for the development of aspirin-exacerbated respiratory disease in smokers. *J Allergy Clin Immunol Pract* 2018;6:116-125 e113.
81. Buchheit KM, Cahill KN, Katz HR, et al. Thymic stromal lymphopoietin controls prostaglandin D2 generation in patients with aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2016;137:1566-1576 e1565.
82. Pan D, Buchheit KM, Samuchiwal SK, et al. COX-1 mediates IL-33-induced extracellular signal-regulated kinase activation in mast cells: Implications for aspirin sensitivity. *J Allergy Clin Immunol* 2019;143:1047-1057 e1048.
83. White AA, Doherty TA. Role of group 2 innate lymphocytes in aspirin-exacerbated respiratory disease pathogenesis. *Am J Rhinol Allergy* 2018;32:7-11.
84. Comhair SAA, Bochenek G, Baicker-McKee S, et al. The utility of biomarkers in diagnosis of aspirin exacerbated respiratory disease. *Respir Res* 2018;19:210.
85. Sladek K, Szczeklik A. Cysteinyl leukotrienes overproduction and mast cell activation in aspirin-provoked bronchospasm in asthma. *Eur Respir J* 1993;6:391-399.
86. Choi GS, Kim JH, Shin YS, Ye YM, Kim SH, Park HS. Eosinophil activation and novel mediators in the aspirin-induced nasal response in AERD. *Clin Exp Allergy* 2013;43:730-740.
87. Mita H, Higashi N, Taniguchi M, Higashi A, Kawagishi Y, Akiyama K. Urinary 3-bromotyrosine and 3-chlorotyrosine concentrations in asthmatic patients: lack of increase in 3-bromotyrosine concentration in urine and plasma proteins in aspirin-induced asthma after intravenous aspirin challenge. *Clin Exp Allergy* 2004;34:931-938.
88. Laidlaw TM, Kidder MS, Bhattacharyya N, et al. Cysteinyl leukotriene overproduction in aspirin-exacerbated respiratory disease is driven by platelet-adherent leukocytes. *Blood* 2012;119:3790-3798.
89. Mitsui C, Kajiwara K, Hayashi H, et al. Platelet activation markers overexpressed specifically in patients with aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2016;137:400-411.

90. Laidlaw TM, Cahill KN, Cardet JC, et al. A trial of type 12 purinergic (P2Y₁₂) receptor inhibition with prasugrel identifies a potentially distinct endotype of patients with aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2019;143:316-324 e317.
91. Cahill KN, Bensko JC, Boyce JA, Laidlaw TM. Prostaglandin D₂: a dominant mediator of aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2015;135:245-252.
92. Cahill KN, Murphy K, Singer J, Israel E, Boyce JA, Laidlaw TM. Plasma tryptase elevation during aspirin-induced reactions in aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2019;143:799-803 e792.
93. Isogai S, Niwa Y, Yatsuya H, et al. Increased airway hyperresponsiveness to adenosine in patients with aspirin intolerant asthma. *Allergol Int* 2017;66:360-362.
94. Taniguchi M, Mitsui C, Hayashi H, et al. Aspirin-exacerbated respiratory disease (AERD): Current understanding of AERD. *Allergol Int* 2019;68:289-295.
95. Hayashi H, Fukutomi Y, Mitsui C, et al. Omalizumab for aspirin hypersensitivity and leukotriene overproduction in aspirin-exacerbated respiratory disease. A randomized controlled trial. *Am J Respir Crit Care Med* 2020;201:1488-1498.
96. Tuttle KL, Buchheit KM, Laidlaw TM, Cahill KN. A retrospective analysis of mepolizumab in subjects with aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol Pract* 2018;6:1045-1047.
97. Laidlaw TM, Mullol J, Fan C, et al. Dupilumab improves nasal polyp burden and asthma control in patients with CRSwNP and AERD. *J Allergy Clin Immunol Pract* 2019;7:2462-2465 e2461.
98. Agarwal R, Chakrabarti A, Shah A, et al. Allergic bronchopulmonary aspergillosis: review of literature and proposal of new diagnostic and classification criteria. *Clin Exp Allergy* 2013;43:850-873.
99. Chowdhary A, Agarwal K, Kathuria S, Gaur SN, Randhawa HS, Meis JF. Allergic bronchopulmonary mycosis due to fungi other than *Aspergillus*: a global overview. *Crit Rev Microbiol* 2014;40:30-48.
100. Oshikata C, Watanabe M, Saito A, et al. Allergic bronchopulmonary mycosis caused by *Penicillium luteum*. *Med Mycol Case Rep* 2017;15:9-11.
101. Castanhinha S, Sherburn R, Walker S, et al. Pediatric severe asthma with fungal sensitization is mediated by steroid-resistant IL-33. *J Allergy Clin Immunol* 2015;136:312-322 e317.
102. Oguma T, Taniguchi M, Shimoda T, et al. Allergic bronchopulmonary aspergillosis in Japan: A nationwide survey. *Allergol Int* 2018;67:79-84.

- Accepted Article
103. Fraczek MG, Chishimba L, Niven RM, et al. Corticosteroid treatment is associated with increased filamentous fungal burden in allergic fungal disease. *J Allergy Clin Immunol* 2018;142:407-414.
 104. Watai K, Fukutomi Y, Hayashi H, et al. De novo sensitization to *Aspergillus fumigatus* in adult asthma over a 10-year observation period. *Allergy* 2018;73:2385-2388.
 105. Ishiguro T, Takayanagi N, Kagiya N, Shimizu Y, Yanagisawa T, Sugita Y. Clinical characteristics of biopsy-proven allergic bronchopulmonary mycosis: variety in causative fungi and laboratory findings. *Intern Med* 2014;53:1407-1411.
 106. Bosken CH, Myers JL, Greenberger PA, Katzenstein AL. Pathologic features of allergic bronchopulmonary aspergillosis. *Am J Surg Pathol* 1988;12:216-222.
 107. Hebisawa A, Tamura A, Kurashima A, et al. [Pathologic reconsideration on allergic bronchopulmonary aspergillosis and mycosis]. *Nihon Kokyuki Gakkai Zasshi* 1998;36:330-337.
 108. Dykewicz MS, Rodrigues JM, Slavin RG. Allergic fungal rhinosinusitis. *J Allergy Clin Immunol*. 2018;142:341-351.
 109. Li JX, Fan LC, Li MH, Cao WJ, Xu JF. Beneficial effects of omalizumab therapy in allergic bronchopulmonary aspergillosis: A synthesis review of published literature. *Respir Med* 2017;122:33-42.
 110. Tillie-Leblond I, Germaud P, Leroyer C, et al. Allergic bronchopulmonary aspergillosis and omalizumab. *Allergy* 2011;66:1254-1256.
 111. Tomomatsu K, Oguma T, Baba T, et al. Effectiveness and safety of omalizumab in patients with allergic bronchopulmonary aspergillosis complicated by chronic bacterial infection in the airways. *Int Arch Allergy Immunol* 2020;181:499-506.
 112. Voskamp AL, Gillman A, Symons K, et al. Clinical efficacy and immunologic effects of omalizumab in allergic bronchopulmonary aspergillosis *J Allergy Clin Immunol Pract* 2015;3:192-199.
 113. Schleich F, Vaia ES, Pilette C, et al. Mepolizumab for allergic bronchopulmonary aspergillosis: Report of 20 cases from the Belgian Severe Asthma Registry and review of the literature. *J Allergy Clin Immunol Pract* 2020;8:2412-2413 e2412.
 114. Tomomatsu K, Sugino Y, Okada N, Tanaka J, Oguma T, Asano K. Rapid clearance of mepolizumab-resistant bronchial mucus plugs in allergic bronchopulmonary aspergillosis with benralizumab treatment. [published ahead of print Apr 1, 2020] *Allergol Int* doi: 10.1016/j.alit.2020.03.003.

115. Ramonell RP, Lee FE, Swenson C, Kuruvilla M. Dupilumab treatment for allergic bronchopulmonary aspergillosis: A case series. *J Allergy Clin Immunol Pract* 2020;8:742-743.
116. Fokkens WJ, Lund VJ, Hopkins C, et al. European position paper on rhinosinusitis and nasal polyps 2020. *Rhinology* 2020;58:1-464.
117. Fujieda S, Imoto Y, Kato Y, et al. Eosinophilic chronic rhinosinusitis. *Allergol Int* 2019;68:403-412.
118. Tokunaga T, Sakashita M, Haruna T, et al. Novel scoring system and algorithm for classifying chronic rhinosinusitis: the JESREC Study. *Allergy* 2015;70:995-1003.
119. Stevens WW, Peters AT, Tan BK, et al. Associations between inflammatory endotypes and clinical presentations in chronic rhinosinusitis. *J Allergy Clin Immunol Pract* 2019;7:2812-2820 e2813.
120. Takabayashi T, Schleimer RP. Formation of nasal polyps: The roles of innate type 2 inflammation and deposition of fibrin. *J Allergy Clin Immunol* 2020;145:740-750.
121. Takabayashi T, Kato A, Peters AT, et al. Excessive fibrin deposition in nasal polyps caused by fibrinolytic impairment through reduction of tissue plasminogen activator expression. *Am J Respir Crit Care Med* 2013;187:49-57.
122. Takabayashi T, Tanaka Y, Susuki D, et al. Increased expression of L-plastin in nasal polyp of patients with nonsteroidal anti-inflammatory drug-exacerbated respiratory disease. *Allergy* 2019;74:1307-1316.
123. Shimizu S, Gabazza EC, Ogawa T, et al. Role of thrombin in chronic rhinosinusitis-associated tissue remodeling. *Am J Rhinol Allergy* 2011;25:7-11.
124. Takabayashi T, Kato A, Peters AT, et al. Increased expression of factor XIII-A in patients with chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol* 2013;132:584-592 e584.
125. Imoto Y, Kato A, Takabayashi T, et al. Increased thrombin-activatable fibrinolysis inhibitor levels in patients with chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol* 2019;144:1566-1574 e1566.
126. Kidoguchi M, Noguchi E, Nakamura T, et al. DNA methylation of proximal PLAT promoter in chronic rhinosinusitis with nasal polyps. *Am J Rhinol Allergy* 2018;32:374-379.
127. Imoto Y, Kato A, Takabayashi T, et al. Short-chain fatty acids induce tissue plasminogen activator in airway epithelial cells via GPR41&43. *Clin Exp Allergy* 2018;48:544-554.
128. Ninomiya T, Noguchi E, Haruna T, et al. Periostin as a novel biomarker for postoperative recurrence of chronic rhinosinitis with nasal polyps. *Sci Rep* 2018;8:11450.

129. Kato Y, Takabayashi T, Sakashita M, et al. Expression and functional analysis of CST1 in intractable nasal polyps. *Am J Respir Cell Mol Biol* 2018;59:448-457.
130. Bachert C, Han JK, Desrosiers M, et al. Efficacy and safety of dupilumab in patients with severe chronic rhinosinusitis with nasal polyps (LIBERTY NP SINUS-24 and LIBERTY NP SINUS-52): results from two multicentre, randomised, double-blind, placebo-controlled, parallel-group phase 3 trials. *Lancet* 2019;394:1638-1650.
131. Fokkens WJ, Lund V, Bachert C, et al. EUFOREA consensus on biologics for CRSwNP with or without asthma. *Allergy* 2019;74:2312-2319.
132. Matsunaga K, Katoh N, Fujieda S, Izuhara K, Oishi K. Dupilumab: Basic aspects and applications to allergic diseases. *Allergol Int* 2020;69:187-196.
133. Numata T, Nakayama K, Utsumi H, et al. Efficacy of mepolizumab for patients with severe asthma and eosinophilic chronic rhinosinusitis. *BMC Pulm Med* 2019;19:176.
134. Tsurumaki H, Matsuyama T, Ezawa K, et al. Rapid effect of benralizumab for hypereosinophilia in a case of severe asthma with eosinophilic chronic rhinosinusitis. *Medicina (Kaunas)* 2019;55.
135. Gevaert P, Omachi TA, Corren J, et al. Efficacy and safety of omalizumab in nasal polyposis: 2 randomized phase 3 trials. [published ahead of print Jun 7, 2020] *J Allergy Clin Immunol*. doi: 10.1016/j.jaci.2020.05.032.
136. Cottin V, Bel E, Bottero P, et al. Respiratory manifestations of eosinophilic granulomatosis with polyangiitis (Churg-Strauss). *Eur Respir J* 2016;48:1429-1441.
137. Furuta S, Iwamoto T, Nakajima H. Update on eosinophilic granulomatosis with polyangiitis. *Allergol Int* 2019;68:430-436.
138. Groh M, Pagnoux C, Baldini C, et al. Eosinophilic granulomatosis with polyangiitis (Churg-Strauss) (EGPA) Consensus Task Force recommendations for evaluation and management. *Eur J Intern Med* 2015;26:545-553.
139. Vaglio A, Buzio C, Zwerina J. Eosinophilic granulomatosis with polyangiitis (Churg-Strauss): state of the art. *Allergy* 2013;68:261-273.
140. Berti A, Cornec D, Casal Moura M, et al. Eosinophilic granulomatosis with polyangiitis: clinical predictors of long-term asthma severity. *Chest* 2020;157:1086-1099.
141. Chang HC, Chou PC, Lai CY, Tsai HH. Antineutrophil cytoplasmic antibodies and organ-specific manifestations in eosinophilic granulomatosis with polyangiitis: A systematic review and meta-analysis. [published online ahead of print, 2020 Aug 6] *J Allergy Clin Immunol Pract*. doi:10.1016/j.jaip.2020.07.038

142. Tsurikisawa N, Oshikata C, Watanabe M, Tsuburai T, Kaneko T, Saito H. Innate immune response reflects disease activity in eosinophilic granulomatosis with polyangiitis. *Clin Exp Allergy* 2018;48:1305-1316.
143. Vega Villanueva KL, Espinoza LR. Eosinophilic vasculitis. *Curr Rheumatol Rep* 2020;22:5.
144. Dong ZM, Lin E, Wechsler ME, et al. Pulmonary eosinophilic granulomatosis with polyangiitis has IgG4 plasma cells and immunoregulatory features. *Am J Pathol* 2020;190:1438-1448.
145. Jakiela B, Sanak M, Szczeklik W, et al. Both Th2 and Th17 responses are involved in the pathogenesis of Churg-Strauss syndrome. *Clin Exp Rheumatol* 2011;29:S23-34.
146. Jakiela B, Szczeklik W, Plutecka H, et al. Increased production of IL-5 and dominant Th2-type response in airways of Churg-Strauss syndrome patients. *Rheumatology (Oxford)* 2012;51:1887-1893.
147. Hellmich B, Csernok E, Gross WL. Proinflammatory cytokines and autoimmunity in Churg-Strauss syndrome. *Ann N Y Acad Sci* 2005;1051:121-131.
148. Higashi N, Mita H, Taniguchi M, et al. Urinary eicosanoid and tyrosine derivative concentrations in patients with vasculitides. *J Allergy Clin Immunol* 2004;114:1353-1358.
149. Marvisi C, Sinico RA, Salvarani C, et al. New perspectives in eosinophilic granulomatosis with polyangiitis (EGPA): report of the first meeting of the European EGPA Study Group. *Intern Emerg Med* 2019;14:1193-1197.
150. Szczeklik W, Sanak M, Mastalerz L, et al. 12-hydroxy-eicosatetraenoic acid (12-HETE): a biomarker of Churg-Strauss syndrome. *Clin Exp Allergy* 2012;42:513-522.
151. Wechsler ME, Akuthota P, Jayne D, et al. Mepolizumab or Placebo for Eosinophilic Granulomatosis with Polyangiitis. *N Engl J Med* 2017;376:1921-1932.
152. Basta F, Mazzuca C, Nucera E, Schiavino D, Afeltra A, Antonelli Incalzi R. Omalizumab in eosinophilic granulomatosis with polyangiitis: friend or foe? A systematic literature review. *Clin Exp Rheumatol* 2020;38 Suppl 124:214-220.
153. Hong H, Liao S, Chen F, Yang Q, Wang DY. Role of IL-25, IL-33, and TSLP in triggering united airway diseases toward type 2 inflammation. [published online ahead of print, 2020 Aug 1] *Allergy*. doi:10.1111/all.14526
154. Nakajima S, Kabata H, Kabashima K, Asano K. Anti-TSLP antibodies: Targeting a master regulator of type 2 immune responses. *Allergol Int.* 2020;69:197-203.

Table 1. Genetic Loci associated with adult asthma-related phenotypes from at least two GWASs among five GWASs

Locus	Reported or mapped genes	Ref.38	Ref.41	Ref.42	Ref.40	Ref.39	
		Adult-onset asthma			EGPA	NP	CRS
1q24.2 [†]	<i>CD247, POU2F1</i> <i>GPA33</i>						
1q31.3	<i>PTPRC</i>						
2q12.1	<i>IL1RL1, IL1RL2, IL18R1, SLC9A4,</i>						
2q13	<i>ACOXL, BCL2L11</i>						
2q22.3	<i>TEX41, PABPC1P2, ACVR2A, RPL6P5</i>						
2q37.3	<i>D2HGDH, ING5, GAL3ST2</i>						
3p22.3	<i>GLB1</i>						
3q26.32	<i>TBL1XR1</i>						
5q22.1	<i>SLC25A46, TSLP, WDR36</i>						
5q31.1 [†]	<i>C5orf56, SLC22A5, IRF1</i> <i>SLC22A4</i>						
5q31.3	<i>NDFIP1</i>						
6p21.3 [‡]	<i>HLA-DQA1, HLA-DQB, HLA-B, HLA-C</i>						
6q15	<i>BACH2, MAP3K7</i>						
8q21.13	<i>TPD52, MIR5708, ZBTB10</i>						
9q24.1	<i>RANBP6, IL33, GTF3AP1</i>						
10p14	<i>GATA3, SFTA1P, CELF2</i>						
10p15.1	<i>IL2RA</i>						
11p15.5	<i>MUC2, MUC5B, MUC6, MUC5AC</i>						
11q12.2	<i>MYRF, FADS2, FADS1, FADS3</i>						
11q13.5	<i>WNT11, LRRC32, EMSY</i>						
11q23.1	<i>LAYN, SIK2</i>						
12q13.11	<i>HDAC7, SLC48A1, VDR</i>						
12q13.2	<i>RPS26, ERBB3, RAB5B, CDK2, SUOX</i>						
12q13.3	<i>STAT6, NAB2, LRP1</i>						
13q14.11	<i>COG6</i>						
15q22.2	<i>RORA</i>						
15q22.33	<i>SMAD3, SMAD6, AAGAB</i>						
16p12.1	<i>IL4R</i>						
16p13.13	<i>CLEC16A, CIITA, RMI2</i>						
17q21.33	<i>ZNF652, PHB</i>						
19q13.11	<i>SLC7A10, CEBPA, LRP3, CEBPA</i>						
21q22.12	<i>RUNX1, SETD4</i>						

[†]These reported genes are located within 500 kb.

[‡]Representative genes are shown at the HLA locus.

The reported or mapped genes of GWASs of adult-onset asthma were obtained from GWAS Catalog (<https://www.ebi.ac.uk/gwas/>).

Table 2. Clinical characteristics and treatment responses to biologics of adult-onset eosinophilic airway diseases

	Adult-onset eosinophilic severe asthma	N-ERD	ABPA	eosinophilic CRS	EGPA
Prevalence in adult asthma	21-48%	5-10%	2.5%	3-10%	0.6%
Male:Female	2:3	1:2	1:1	2:1	1:2
Age of onset, years	30 - 46	35 - 36	25 - 55	30 - 50	56
at ≤12 years	N/A	rare	rare	rare	rare
at ≥ 60 years	not rare	rare	not rare	rare	not rare
Comorbidities					
Asthma	(100%)	100%	67-100%	24-67%	90-100%
ECRS	10-85%	90-100%	rare	100%	70-80%
Atopy	52-80%	34-73%	90-100%	30-40%	38%
Disease-specific clinical characteristics	Rapid decline of pulmonary functions	NSAIDs intolerance	Mucus plugs	Nasal polyps Viscous colloidal	Systemic vasculitis Granulomatosis
Mucus plugs in the airway lumen	+	±	++	++	±
Biomarkers					
Peripheral blood eosinophils	↑ ↑	↑	↑ ↑ ↑	↑	↑ ↑ ↑
Serum total IgE, IU/mL		100-300	1,000-10,000	200-300	100-300
FeNO	↑	↑	↑	↑, nasal NO ↓	↑
Serum periostin	↑	↑	↑ ↑	↑	↑ →
Urinary LTE4		↑ ↑ ↑		↑	↑ ↑
EETs/EETosis	+		++	++	±
	(lumen/tissue)		(lumen)	(lumen/tissue)	(tissue)
Charcot-Leyden crystals	+	-	++	++	±
	(lumen/tissue)		(lumen)	(lumen/tissue)	(tissue)

Other biomarkers

Mast cell/
platelet
activation
markers

CST-1
eotaxin-3
IgG4

ANCA
eotaxin-3
soluble IL-2R

Responses to biologics

anti-IgE Ab	+	++	+	+	+
anti-IL-5 Ab/anti-IL-5R α Ab	++		++	+	++
anti-IL-4R α Ab	++	+		++	
anti-TSLP Ab	++				

Abbreviations: ABPA, allergic bronchopulmonary aspergillosis; CRS, chronic rhinosinusitis; EETs/EETosis, eosinophil extracellular traps/eosinophil ETosis; EGPA, eosinophilic granulomatosis with polyangiitis; IgE, Immunoglobulin E; LTE₄, leukotriene E₄; N/A, not applicable; NSAID, nonsteroidal anti-inflammatory drug; N-ERD, NSAID-exacerbated respiratory disease.

Figure legends

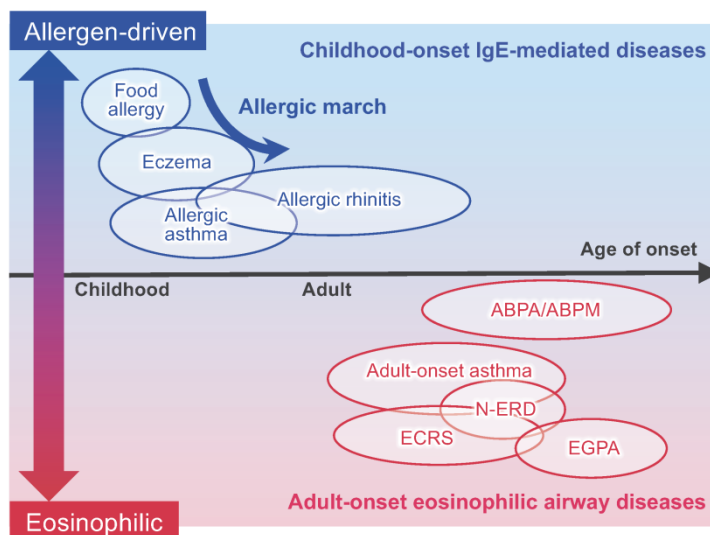
Figure 1. Childhood-onset IgE-mediated diseases and adult-onset eosinophilic airway diseases

Childhood-onset allergic diseases such as food allergy, eczema, allergic asthma and rhinitis are mediated by allergen-specific IgE. These diseases often develop consecutively in a single patient (allergic march). In contrast, adult-onset allergic airway diseases including asthma, eosinophilic CRS, nonsteroidal anti-inflammatory drug-exacerbated respiratory disease (N-ERD), allergic bronchopulmonary aspergillosis/mycosis (ABPA/ABPM), and eosinophilic granulomatosis with polyangiitis (EGPA) are not mediated by IgE, but often accompanied by prominent peripheral blood eosinophilia, and develops simultaneously or consecutively.

Figure 2. Eosinophil-mediated chronic airway inflammation.

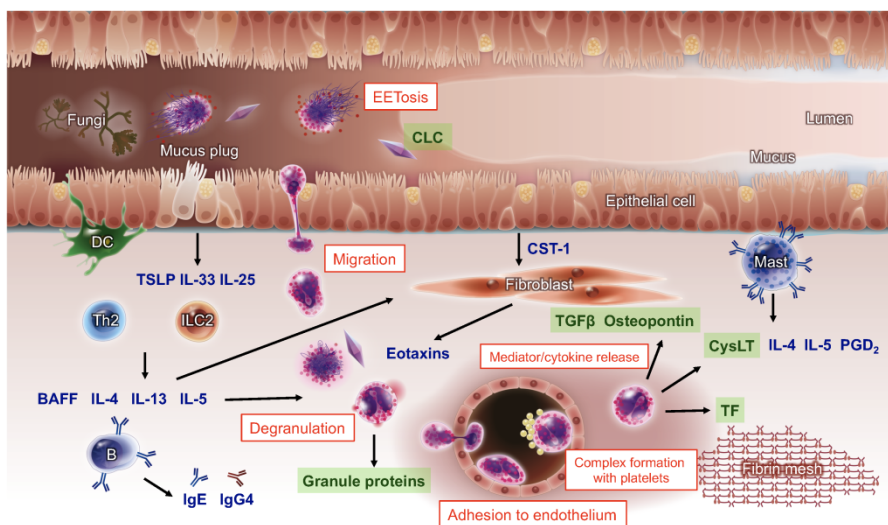
In addition to allergen-induced Th2 cell activation, epithelial alarmin cytokines IL-25, IL-33, and TSLP act to induce the local production of IL-5, which serves as the key molecule for eosinophil proliferation. IL-4 and IL-13 play a role in eosinophil-selective extravasation through the induction of adhesion molecule expression on the endothelium. IL-4 and IL-13 further contribute to tissue eosinophilia through production of eotaxins (CCL11/eotaxin-1, CCL24/eotaxin-2, CCL26/eotaxin-3) from fibroblasts and epithelial cells. IL-5 and various local mediators activate eosinophils to release toxic granule proteins through secretion mechanism and cytolytic EETosis. Transepithelial eosinophil migration followed by EETosis, and Charcot-Leyden crystal (CLC) formation, cause fixed airway obstruction in several airway diseases such as ABPM characterised by highly viscous allergic (eosinophilic) mucin harbouring fungal hyphae in the central bronchi. In nasal polyps, CST-1 elicits type 2 immunity and eosinophil-derived tissue factor (TF) activates coagulation cascade together with diminished fibrinolysis system induced excessive fibrin deposition. The complex formation of eosinophils and platelets lead to overproduction of cysLT in N-ERD. Eosinophils also serve as a major source of cytokines and mediators involved in tissue remodelling.

Fig.1



all_14620_f1.tiff

Fig.2



all_14620_f2.tiff

Supplementary information

Adult-onset eosinophilic airway diseases

Koichiro Asano, Shigeharu Ueki, Mayumi Tamari, Yoshimasa Imoto, Shigeharu Fujieda, and Masami Taniguchi

Table E1.

Recent GWASs of inflammatory airway diseases relating to adult-onset eosinophilic airway diseases

Figure E1.

Diagnostic algorithm for eosinophilic CRS severity

Table E1. Recent GWASs of inflammatory airway diseases relating to adult-onset eosinophilic airway diseases

Study phenotype and population			
Ref. 38) Age of asthma onset: UK biobank cohort, European			
	Sub-phenotype	Cases	Controls
1st GWAS	Childhood-onset asthma (COA) (onset: 0-19 y)	13,962	300,671
	Adult-onset asthma (AOA) (onset: 20-60 y)	26,582	300,671
Validation	Childhood-onset asthma (COA) (onset: 0-19 y)	31,759	214,890
	Adult-onset asthma (AOA) (onset: 20-60 y)	16,297	217,711
	Childhood-onset asthma (n=31,002) vs Adult-onset asthma (n=16,297)		
Methods	GWAS, expression quantitative trait loci (eQTL) and non-synonymous SNPs to predict target genes		
Main results	The variance in disease liability explained by common variants: COA (h²_g=25.6%) > AOA (h²_g=10.6%)		
	The genetic correlation between COA and AOA: r_g=0.67		
	GWAS loci: 123 independent associations for COA and 56 for AOA (37 overlapped)		
	:of these, 98 and 34, respectively, were replicated in an independent study		
	The GWAS found 28 novel associations, 5 COA-specific risk variants, and 3 variants that are stronger risk factors for AOA		
	Variants associated with obesity and smoking had a stronger effect to the risk of AOA than to the risk of COA		
	The study identified 109 likely target genes on the basis of correlated eQTL		
Ref. 41) Age of asthma onset: UK biobank cohort, British white individuals			
	Sub-phenotype	Cases	Controls
	Childhood-onset asthma (onset: 0-12 y) (1)	9,433	318,237
	Adult-onset asthma (onset: 26-65 y) (2)	21,564	318,237
	Adolescent or young adult onset (onset: 12-25 y) (3)	6,849	
	Age of onset as a quantitative trait (1)+(2)+(3)	37,846	
Methods	GWAS, Enrichment studies to determine the tissues in which genes at GWAS loci were most highly expressed		
	Transcriptome-wide gene-based test (PrediXcan) to identify candidate risk genes		
Main results	The study identified 61 independent asthma loci: 23 childhood-onset specific loci, 1 adult-onset specific loci, and 37 shared loci.		
	19 loci were associated with age of asthma onset		
	Most highly expressed tissues of genes at the loci	Childhood onset-specific loci: Skin, blood, and small intestine	
		Adult onset-specific loci: Lung, blood, small intestine, and spleen	
	113 unique candidate genes at 22 of the 61 GWAS loci were identified by PrediXcan		
	SNP-based heritability estimates: childhood-onset asthma (0.327) > adult-onset asthma (0.098)		
Ref. 42) Obesity-related traits and asthma: UK biobank cohort, European			
	Sub-phenotype	Cases	Controls
	Later-onset asthma (onset: ≥16 y)	34,286	402,763
	Early onset asthma (onset: <16 y)	13,764	402,763
	Non-atopic asthma	29,605	319,321
	Atopic asthma	24,600	432,368
Methods	GWAS, Obese versus lean mouse RNA sequencing and RT-PCR experiments		
	Mendelian randomization analysis		
Main results	Positive genetic correlation between BMI and later-onset asthma (onset: ≥16 y) (Rg = 0.25)		
	Mendelian randomisation suggested BMI causally increasing asthma risk		
	Cross-trait GWAS identified 34 shared loci among 3 obesity-related traits and 2 asthma subtypes		

Ref. 40) Eosinophilic granulomatosis with polyangiitis (EGPA): European cohorts

	Sub-phenotype	Cases	Controls
Primary cohort	Total	534	6,688
	ANCA negative	352	
	MPO positive	159	
Secondary	Total	142	125
	MPO positive	43	

Methods GWAS, conditional false discovery rate (cFDR) analysis, Mendelian randomisation analysis

Main results The study identified 4 loci through conventional GWAS and 4 associations through cFDR approach

Mendelian randomisation suggested a primary tendency to eosinophilia contributes to EGPA susceptibility

EGPA comprises 2 genetically clinically distinct syndromes, MPO+ ANCA EGPA and ANCA-negative EGPA

Four candidate genes (*IL5*, *TSLP*, *BCL2*, *CDK6*) are targets of therapies in development, supporting their exploration in EGPA

Ref. 39) Nasal polyps (NP) and CRS: UK biobank cohort and Iceland cohort, European

	Sub-phenotype	Cases	Controls
	NP Iceland	1,175	309,305
	NP UK biobank	3,191	405,379
	CRS Iceland	3,188	353,939
	CRS UK biobank	2,420	406,147

Methods GWAS

Main results NP meta-analysis identified 10 loci (*ALOX15*, *HLA*, *IL18R1-IL1RL1*, *IL33*, *FOXP1*, *CYP2S1*, *SLC22A4*, *MYRF*, *TSLP*, *10p14*)

CRS meta-analysis identified 2 loci (*ALOX15*, *HLA*)

ALOX15 (p.Thr560Met) was reported to cause near total loss of 15-LO enzymatic activity

ALOX15 (p.Thr560Met) confers large genome-wide significant protection against NP (OR=0.32) and CRS (OR=0.64)

Abbreviation: EGPA, eosinophilic granulomatosis with polyangiitis.

Figure E1. Diagnostic algorithm for eosinophilic CRS severity

Patients with CRS with nasal polyp would be classified into four groups by JESREC score: blood eosinophilia, ethmoid-dominant shadow in CT, and comorbidity (asthma and nonsteroidal anti-inflammatory drug-exacerbated respiratory disease [N-ERD]).

Fig.E1

