Advances in Cellular and Molecular Otolaryngology

COACTION PUBLISHING

REVIEW ARTICLE

Biological basis and clinical implications of immunological molecules involved in eosinophilic inflammation in allergic rhinitis, chronic rhinosinusitis, and asthma

Shin Kariya*, Mitsuhiro Okano and Kazunori Nishizaki

Department of Otolaryngology–Head and Neck Surgery, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama, Japan

Eosinophils are specialized and multifunctional immune cells that are involved in both protection and pathology in human health and disease. Eosinophils have been thought to play an important role in parasitic infection, and defense against parasites is one of the main functions of eosinophils. Eosinophils also have damaging effects on the pathogenesis of numerous diseases, especially allergic disorders. A large number of inflammatory mediators including cytokines, chemokines, and adhesion molecules are associated with the development and chemotaxis of eosinophils. Allergic rhinitis and asthma are typical respiratory diseases with eosinophilic inflammation, and eosinophils are also involved in the onset and development of some types of chronic rhinosinusitis. The purpose of this review is to summarize the physiology of eosinophils and to discuss potential therapeutic targets related to eosinophils in allergic rhinitis, chronic rhinosinusitis, and asthma.

Keywords: *eosinophil*; *rhinitis*; *rhinosinusitis*; *sinusitis*; *asthma*; *airway*; *cytokine*; *chemokine*; *interleukin*; *receptor*

*Correspondence to: Shin Kariya, Department of Otolaryngology–Head and Neck Surgery, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, 2-5-1 Shikata-cho, Kita-ku, Okayama, 700-8558, Japan, Email: skariya@cc.okayama-u.ac.jp

Received: 10 November 2014; Revised: 10 January 2015; Accepted: 14 January 2015; Published: 17 February 2015

Eosinophils are a type of white blood cell produced in the bone marrow and are particularly involved in immune responses to allergic inflammation and parasitic infection. Since the discovery regarding the capacity of causing tissue damage by eosinophil-derived granule proteins, eosinophils have become recognized as one of the main players in the immune system. Eosinophils also have the ability to synthesize a variety of inflammatory mediators and play an important role in homeostatic mechanisms.

Eosinophils play an important role in pathophysiology in upper and lower respiratory diseases including allergic rhinitis, chronic rhinosinusitis, and asthma (1–5). Allergic rhinitis is a typical type 1 [immunoglobulin (Ig) E-mediated] hypersensitivity immune response to environmental allergens. Eosinophilic infiltration of the nasal mucosa and subepithelial edema are characteristic features of allergic rhinitis. The presence of eosinophils in nasal secretions is an important finding in the diagnosis of allergic rhinitis. Chronic rhinosinusitis is usually classified into three categories: chronic rhinosinusitis without nasal polyposis (CRSsNP), chronic rhinosinusitis with nasal polyposis (CRSwNP), and allergic fungal rhinosinusitis (AFRS) (3, 6, 7). Eosinophilia is more commonly associated with CRSwNP than with CRSsNP, and recent studies show that CRSwNP and CRSsNP have disparate clinical features based on distinct inflammatory pathways, cytokine profiles, and different tissue remodeling (8–19). AFRS is a non-invasive form of fungal rhinosinusitis that represents an allergic hypersensitivity disorder (IgE mediated in some cases) (20). The presence of allergic mucin in the sinonasal cavity containing fungal hyphae and a large number of degranulating eosinophils is a significant finding in AFRS (21, 22). Asthma is a common but complex disease involving type I hypersensitivity reactions (23). Eosinophils and Th2 cytokines such as interleukin (IL)-4, IL-5, and IL-13 play an important role in the pathophysiology of asthma. Chemokine (C-C motif) ligand (CCL)

Citation: Advances in Cellular and Molecular Otolaryngology 2015, **3**: 26601 - http://dx.doi.org/10.3402/acmo.v3.26601

11 (also known as eotaxin-1), CCL24 (also known as eotaxin-2), and CCL26 (also known as eotaxin-3) derived from epithelial cells mediate eosinophil recruitment into the asthmatic lung under the influence of cytokines such as IL-13 (24).

The similarity of anatomical and immunological findings in the upper and lower airway has been reported, and numerous studies show a close relationship between sinonasal diseases and lower respiratory diseases (25–31). The purpose of this review is to summarize the role of eosinophils in allergic rhinitis, chronic rhinosinusitis, and asthma, as well as to describe recent advances in the therapeutic targets related to eosinophils in the management of allergic rhinitis, chronic rhinosinusitis, and asthma.

Differentiation of eosinophils

Eosinophils are produced in bone marrow from pluripotent stem cells. In regulating eosinophil development, IL-3, IL-5, granulocyte–macrophage colony-stimulating factor (GM-CSF), and several transcription factors including GATA-1 (a zinc finger family member), PU.1 (an Ets family member), and CCAAT/enhancer-binding protein (c/EBP) family members (c/EBPα, c/EBPβ, and c/EBPε) are particularly important factors, and synergistically regulate activity (32, 33). In these transcription factors, GATA-1 is clearly the most important in specifying eosinophil lineage. The experimental study reveals that mice with a targeted deletion of the high-affinity GATA-binding site present in the GATA-1 promoter gene show loss of the eosinophil lineage (34); these findings are supported by eosinophil differentiation experiments *in vitro* (35, 36). IL-3 and GM-CSF also induce the proliferation of neutrophils and basophils and are not relatively specific for the development of eosinophils. However, IL-5 potently and specifically stimulates eosinophil production in bone marrow (37).

Adhesion and migration of eosinophils

Eosinophil migration from bone marrow into circulating blood is primarily regulated by IL-5 (38). Under normal conditions, most eosinophils in peripheral organs are found in the gastrointestinal tract except the esophagus, mammary gland, uterus, and thymus. Among these organs, the gastrointestinal eosinophil is predominant (39). In healthy individuals, eosinophils in the gastrointestinal tract are regulated by the interaction between CCL11 and its receptor [CCR3, chemokine (C-C motif) receptor 3] and are present independent of adaptive immunity and enteric flora (40, 41). Under baseline condition, eosinophils in the thymus, mammary gland, and uterus are also controlled by CCL11 (42).

Several molecules, including a number of cytokines (IL-4, IL-5, and IL-13), adhesion molecules (β1-, β2-, and β7-integrins), RANTES (regulated on activation, normal T-cell expressed and secreted, also known as CCL5), eotaxins, intercellular adhesion molecule 1 (ICAM-1, also known as CD54), vascular cell adhesion molecule 1 (VCAM-1, also known as CD106), and mucosal addressin cell adhesion molecule 1 (MadCAM-1) are involved in the trafficking of eosinophils into inflammatory sites and adhesion of eosinophils in endothelial cells (43–49). In addition, a recent study shows that the CD2 subsets of immunoglobulin superfamily co receptors (CD48/2B4) have an important role in human eosinophil adhesion and intercellular adhesion, and CD48 is a useful marker for the severity of eosinophilic inflammation (50). The eotaxin/CCR3 axis has a central regulatory role in allergic airway inflammation (51, 52). Tissue eosinophils can likely survive for at least two weeks based on *in vitro* observations (44). Of the cytokines implicated in modulating leukocyte recruitment, only IL-5 and the eotaxins selectively regulate eosinophil trafficking (53).

Eosinophil activation

Human eosinophils are activated by many different molecules including IL-3, IL-4, IL-5, IL-13, and GM-CSF (54, 55). A Th1-type cytokine, interferon-gamma (IFN-γ), and cytokines derived from the epithelium, such as IL-33 and thymic stromal lymphopoietin (TSLP), also stimulate human eosinophils (56–59). In addition to endogenous receptors, human eosinophils also express receptors for pathogen-associated molecular patterns (PAMPs) and danger-associated molecular patterns (DAMPs), including the family of Toll-like receptors (TLRs), nucleotide-binding oligomerization domain (NOD)-like receptors (NLRs: NOD1 and NOD2), retinoic acid-inducible gene I-like receptor (RIG-I), C-type lectin receptor (Dectin-1), and the receptor for advanced glycation end products (RAGE) (60, 61). Human eosinophils are reported to express TLR-1, 2, 3, 4, 5, 6, 7, 9, and 10. These TLRs recognize a wide variety of products that are expressed or produced by microbes (62–67). For example, ligands for TLR-2 (peptidoglycan), TLR-5 (flagellin), and TLR-7 [imiquimod (R837)] induce eosinophil activation and the release of inflammatory mediators (62, 68). Proteases are frequently found in microbes and allergens, and human eosinophils are capable of recognizing proteases including serine protease, aspartate protease, and cysteine protease. Eosinophils activated by proteases demonstrate the active release of pro-inflammatory mediators (69–72). The role of fungi in the development of asthma is well known, and β-glucan, a major fungal cell wall component, activates eosinophils via the β2 integrin molecule (CD18) pathway (33, 73). Human eosinophils also recognize damaged tissue/cell components and endogenous molecules induced from tissue injury, including uric acid, adenosine triphosphate (ATP), high mobility group box (HMGB)-1, and S100 calcium-binding protein family members, resulting in tissue homeostasis (74–76).

Citation: Advances in Cellular and Molecular Otolaryngology 2015, **3**: 26601 - http://dx.doi.org/10.3402/acmo.v3.26601

Eosinophil granule proteins

Eosinophil cytoplasm contains large specific granules that are the principal identifying feature of eosinophils. Activated eosinophils release granule-stored cationic proteins (MBP, major basic protein; ECP, eosinophil cationic protein; EPO, eosinophil peroxidase; and EDN, eosinophil-derived neurotoxin), which exert a range of biological effects on host cells and microbial targets.

MBP is a small cytotoxic protein and is one of the principal and specific proteins present in eosinophils. It is implicated in many pathological conditions including direct cytotoxic effects, allergic reactions, parasitic infections, and stimulation for a variety of cells to produce inflammatory molecules. MBP is classified as two homologs (MBP-1 and MBP-2) encoded by two different genes. MBP-1 is an arginine- and cysteine-rich polypeptide (13.8 kDa) composed of 117 amino acids. MBP-1 is highly toxic to parasites, bacteria, and mammalian cells by disrupting the lipid bilayer membrane or altering the activity of enzymes within tissues (77, 78). MBP-2 is less potent than MBP-1 in *in vitro* biological activities. However, MBP-2 is present only in eosinophils and may be a useful biomarker for eosinophil-associated diseases (79).

ECP is a basic secretion protein involved in the immune response system. ECP also belongs to the RNase A superfamily and is known as ribonuclease 3 (RNase-3). The cDNA sequence for ECP codes for a preprotein of 160 amino acids and a protein of 133 amino acids. The molecular weight of ECP ranges between 16 and 21.4 kDa (32, 80). ECP has marked toxicity for a variety of helminth parasites, hemoflagellates, bacteria, single-stranded RNA viruses, and host tissues (81). *In vitro*, ECP plays a beneficial role in host defense against single-stranded RNA respiratory syncytial virus (82). ECP is also active against both Gram-negative and Gram-positive strains of bacteria by the mechanism of toxicity involving both the bacterial cell wall and the cytoplasmic membrane (78). The bacterial agglutinating activity of ECP is driven by the formation of amyloid-like aggregates at the bacterial cell surface (83). These recent findings suggest that the amyloidogenic behavior of ECP participates in antibacterial host responses to infection, and that the biophysical property of bactericidal N-terminal peptides of ECP is the novel therapeutic target in the development of antimicrobials (84).

EPO is a two-chain protein and is a major enzyme present in eosinophils. The light chain has a molecular mass of 11–15 kDa and the heavy chain a molecular mass of 50–57 kDa (32). EPO is the abundant cationic matrix protein of the specific granule; human eosinophils have 12 μg of EPO per cell (85). EPO catalyzes the formation of cytotoxic oxidants implicated in asthma, allergic inflammatory disorders, and cancer. Oxidant production begins with the generation of superoxide by the nicotinamide adenine dinucleotide phosphate (NADPH) oxidase enzymatic system, and the superoxide dismutates into hydrogen peroxide (H_2O_2) (86). H_2O_2 is converted by EPO to highly reactive halogenating acids, such as hypobromous acid (HOBr), hypochlorous acid (HOCl), or xanthine oxidase (XO) (87). In addition to the ability to produce oxidants, EPO has some cytotoxic effects as a cationic toxin in parasites and mammalian cells in the absence of H_2O_2 and a halide co-factor. Furthermore, EPO exerts both anti-inflammatory and pro-inflammatory activities (88–91).

EDN, also known as RNase-2, is a member of the ribonuclease A superfamily and is a single-chain polypeptide with an observed molecular mass of 18.6 kDa (92). EDN is localized in the matrix of the secondary granule of the eosinophil, but is also detected in mononuclear cells and neutrophils (93). EDN plays a protective role in respiratory syncytial virus infection in the bronchial tract (94, 95). In addition, EDN induces the migration and maturation of dendritic cells. Recent studies show that EDN is an endogenous ligand of TLR-2 and can activate dendritic cells and monocytes/macrophages by triggering the TLR-2-myeloid differentiation factor 88 (Myd88) signaling pathway, resulting in enhanced antigen-specific Th2-biased immune responses and cytokine production (IL-5, IL-6, IL-10, and IL-13) (96, 97).

Among these eosinophil granule proteins, MBP and ECP were extensively examined. Serum ECP concentration can be used as a marker of local and systemic eosinophil expression (98). MBP and ECP have the clinical importance in monitoring eosinophilic activity in allergic rhinitis, chronic rhinosinusitis, and asthma (99–102).

Eosinophils in innate and adaptive immunity

Human eosinophils have the ability to secrete a large number of important inflammatory and regulatory factors including cytokines, chemokines, and growth factors, and express numerous receptors (Tables 1 and 2) (43, 93, 103–107). Eosinophils modulate the immune response in both the innate or non-specific immune system and the adaptive or specific immune system.

In the innate immunity system, eosinophils primarily have a defensive role against large, non-phagocytosable organisms and fungi by the production of cationic proteins, cytokines, and chemokines. However, the excessive reaction in activated eosinophils sometimes has harmful effects on humans, and direct activation of eosinophils by microbe-derived molecules exacerbates allergic inflammation following bacterial and/or viral infections (38). Innate immunity serves as the first line of defense against infection, and pattern recognition receptors including TLR constitute an important class of immune recognition receptors that can recognize pathogenic molecules. Expression of several TLRs in eosinophils has been reported, and the TLR-7 system may represent an important mechanism for host defense against viral infections (62–65). Proteases

Table 1. Inflammatory molecules and growth factors generated by human eosinophils

Cytokines	Chemokines	Growth factors	Lipid mediators
IL-1α	CCL2 (MCP-1)	CXCL1 (GRO-α)	Leukotriene C_4
IL-1β	CCL3 (MIP-1α)	NGF	Leukotriene D_4
IL-2	CCL5 (RANTES)	PDGF	Leukotriene E_4
IL-3	CCL6 (C10)	SCF	Prostaglandin E_1
IL-4	CCL7 (MCP-3)	EGF	Prostaglandin E_2
IL-5	CCL8 (MCP-2)	APRIL	15-HETE
IL-6	CCL9 (MIP-1γ)	VEGF	Thromboxane B2
IL-9	CCL11 (Eotaxin-1)	HB-EGF-LBP	PAF
IL-10	CCL13 (MCP-4)		
IL-11	CXCL1 (GRO-α)		
IL-12	CXCL5 (ENA-78)		
IL-13	CXCL8 (IL-8)		
IL-16	CXCL9 (MIG)		
IL-17	CXCL10 (IP-10)		
IL-18	CXCL11 (I-TAC)		
IL-22	CXCL12 (SDF-1)		
IL-25			
IFN-γ			
GM-CSF			
SCF			
TGF-α			
TGF-β			
TNF-α			
TNF-β			

IL: interleukin, IFN: interferon, GM-CSF: granulocyte macrophage colony-stimulating factor, SCF: stem cell factor, TGF: transforming growth factor, TNF: tumor necrosis factor, CCL: chemokine (C-C motif) ligand, CXCL: chemokine (C-X-C motif) ligand, MCP: monocytes chemoattractant protein, MIP: macrophage inflammatory protein, RANTES: regulated upon activation, normal T-cell expressed, and secreted, GRO: growth-regulated oncogene, ENA: epithelial-derived neutrophil-activating peptide, MIG: monokine induced by gamma interferon, IP: interferon gamma-induced protein, I-TAC: interferon inducible T-cell alpha chemoattractant, SDF: stromal cell-derived factor, NGF: nerve growth factor, DGF: platelet-derived growth factor, EGF: epidermal growth factor, APRIL: a proliferation-inducing ligand, VEGF: vascular endothelial growth factor, HB-EGF-LBP: heparin-binding epidermal growth factor-like binding protein, 15-HETE: 15-hydroxyeicosatetraenoic acid, PAF: platelet-activating factor.

Table 2. Receptors expressed by human eosinophils for cytokines, chemokines, and lipid mediators

Cytokine receptors	Chemokine receptors	Receptors for lipid mediators
IL-2R (CD25/CD122)	CCR1 (CD191)	$CysLT_1R$
IL-3R (CD123/CD131)	CCR2 (CD192)	$CysLT_2R$
IL-4R (CD124/CD132)	CCR3 (CD193)	Leukotriene B_4R
IL-5R (CD125/CD131)	CCR4 (CD194)	DP1
IL-9R (CD129/CD132)	CCR5 (CD195)	DP2 (CRTH2)
IL-12R	CCR6 (CD196)	Prostaglandin E_2R
IL-13RA1 (CD213a1)	CCR8 (CD198)	PAFR
IL-17A/F R (IL17RA/IL17RC)	CCR9 (CDw199)	fMLPR
IL-23R (IL-23R/IL-12Rβ1)	CXCR2	
IL-27R (CD130/WSX-1)	CXCR3 (CD182, CD183)	
IL-31R (IL-31RA/OSMRβ)	CXCR4	
IL-33R (ST2)		
GM-CSFR (CD116/CD131)		
IFN-γR (CDw119)		
SCFR (c-kit, CD117)		
TNF-αR1 (CD120a)		
TNF-αR2 (CD120b)		
TGF-βR		

R: receptors, CD: cluster of differentiation, IL: interleukin, OSMR: oncostatin M receptor, IFN: interferon, GM-CSF: granulocyte macrophage colony-stimulating factor, SCF: stem cell factor, TNF: tumor necrosis factor, TGF: transforming growth factor, CCR: chemokine (C-C motif) ligand receptor, CXCR: chemokine (C-X-C motif) ligand receptor, CysLT: cysteinyl leukotriene, DP: prostaglandin D2 receptor, CRTH2: chemoattractant-homologous receptor expressed on Th2 cells, PAF: platelet-activating factor, fMLP: *N*-formyl-L-methionyl-L-leucyl-phenylalanine.

from various microbes and allergens, such as house dust mites and fungi, induce production of several inflammatory mediators through a family of G-protein-coupled protease-activated receptors (PAR), and human eosinophils constitutively transcribe mRNA for PAR-2 and PAR-3 (69, 108, 109). Eosinophils have multifunctional β_2 integrin molecules (also known as CD18) that recognize β-glucan, a major cell wall component of fungus, and release their cytotoxic granule proteins into the extracellular milieu and onto the surface of fungal organisms, killing the fungus in a contact-dependent manner (110, 111).

Antibodies are the key components of adaptive immunity, and eosinophils play a significant role in adaptive immune responses thorough Fc receptors including FcαR, FcγR, and FcεR. In addition, a large number of adhesion molecules and receptors are involved in the activation and degranulation of eosinophils (Table 3) (43, 93, 103, 104, 112, 113). The Fc receptor for IgA (FcαR, also known as CD89) is expressed in human eosinophils, and IgA_2 is a highly potent stimulus for eosinophil killing of *Schistosoma mansoni* (114). IgG is also considered to be involved in eosinophil activation via FcγR pathways, both in human and mouse (115, 116). IgG, platelet-activating factor (PAF), and GM-CSF stimulate human eosinophils, and increase eosinophil adhesion by activating integrins. Among these integrins, $\alpha_M\beta_2$ integrin (also known as Mac-1, CR3, or CD11b/CD18) and $\alpha_4\beta_1$ integrin (also known as VLA-4 or CD49d/CD29) play a critical role for eosinophil granule protein release (117–119). IgE plays a critical role in respiratory diseases related to allergic reactions including asthma, allergic rhinitis, and chronic rhinosinusitis. Both the high-affinity

Citation: Advances in Cellular and Molecular Otolaryngology 2015, **3**: 26601 - http://dx.doi.org/10.3402/acmo.v3.26601

Table 3. Adhesion molecules, complement receptors, immunoglobulin (Ig) receptors, and members of Ig superfamily related to human eosinophils

Adhesion molecules	Complement receptors	Ig receptors and members of Ig superfamily
$\alpha_4\beta_7$ integrin (CD49d/Ly69)	CR1 (CD35)	FcαR (CD89)
$\alpha_4\beta_1$ integrin (VLA-4, CD49d/CD29)	CR3 ($\alpha_M\beta_2$ integrin, CD11b/CD18)	FcγRII (CD32)
$\alpha_6\beta_1$ integrin (VLA-6, CD49f/CD29)	CR4 ($\alpha_X\beta_2$ integrin, CD11c/CD18)	FcγRIIIa (CD16a)
$\alpha_D\beta_2$ integrin (CD11d/CD18)	CD103	FcγRIIIb (CD16b)
$\alpha_E\beta_7$ integrin (CD103/Ly69)	C1qR	FcεRI
$\alpha_L\beta_2$ integrin (LFA-1, CD11a/CD18)	C3aR	FcεRII (CD23)
$\alpha_M\beta_2$ integrin (CR3, CD11b/CD18)	C5aR (CD88)	Receptor for IgD
$\alpha_X\beta_2$ integrin (CR4, CD11c/CD18)		Receptor for IgM
LFA-3 (CD58)		Siglec-3 (CD33)
L-selectin (CD62L)		Siglec-7
PSGL-1 (CD162)		Siglec-8
Sialyl-Lewis x (CD15s)		Siglec-10
CD44		ICAM-1 (CD54)
CD156		ICAM-3 (CD50)
CD174		Semaphorin-4D (CD100)
		IGSF2 (CD101)
		HLA class I
		HLA-DR
		CD4
		CD47
		CD48
		CD66
		CD244
		CD300a

CD: cluster of differentiation, VLA: very late antigen, LFA: lymphocyte function-associated antigen, CR: complement receptor, PSGL: P-selectin glyocoprotein ligand, Siglec: sialic acid-binding immunoglobulin-like lectin, ICAM: intercellular adhesion molecule, IGSF2: immunoglobulin superfamily member 2, HLA: human leukocyte antigen.

IgE receptor (FcεRI) and the low-affinity IgE receptor (FcεRII, also known as CD23) are found in eosinophils. FcεRI in eosinophils from patients with eosinophilia induce various functions of eosinophils, including degranulation and parasite cytotoxicity (120).

Therapeutic targets related to eosinophils in allergic rhinitis

Allergic rhinitis is a common disease. Its pathophysiology is based on IgE-mediated type I hypersensitivity. Inhaled allergens binding to dendritic cells and mast cells in the upper airway initiate an immune-inflammatory process in allergic rhinitis. Dendritic cells located in the nasal mucosa capture the allergens and act as antigen-presenting cells. In sensitized individuals, allergens deposited onto the nasal mucosa bind the allergen-specific IgE to the surface of mast cells resulting in rapid release of inflammatory mediators such as histamine (1). Histamine, tumor necrosis factor α (TNF-α), and lipid mediators such as leukotriene C_4 and prostaglandin D_2 contribute to the influx of eosinophils into the upper airway by stimulation of the expression of adhesion molecules (ICAM-1, VCAM-1, and E-selectin) in the vascular endothelium, and induce late allergic responses such as nasal obstruction (1, 121). IL-5 also plays a key role in eosinophilic inflammation in allergic rhinitis through the transendothelial migration of eosinophils from circulating blood to the mucosal tissue in the upper airway and eosinophil survival in tissue.

A number of available therapies are recommended in several guidelines including ARIA (Allergic Rhinitis and its Impact on Asthma) guidelines for a step-wise approach to the management of allergic rhinitis (26). Currently available pharmacological agents, immunotherapy, and other therapeutic options for allergic rhinitis include antihistamines, corticosteroids, antileukotrienes, nasal decongestants, mast cell stabilizers, anticholinergic agents, allergen-specific immunotherapy, monoclonal anti-IgE antibody, capsaicin desensitization, and complementary treatments (2).

Glucocorticoids inhibit the production of cytokines including IL-5 and GM-CSF by glucocorticoid receptor agonism and anti-inflammatory action through multiple effects. Glucocorticoids can shorten the half-life of

eosinophils and reduce eosinophil survival in nasal tissue. Glucocorticoids also reduce nasal lavage eosinophilia. The long-term use of systemic corticosteroids induces adverse effects including inhibition of growth in children, disturbances of metabolism, glaucoma and cataract formation, immunosuppression, skin thinning, behavioral abnormalities, and osteoporosis (122). However, administration of intranasal steroids is relatively safe, and the efficacy of topical steroid therapy exceeds the risk of any systemic side effects (123).

Leukotrienes are a family of inflammatory lipid mediators synthesized from arachidonic acid in macrophages, mast cells, and eosinophils. Among the leukotrienes, leukotriene C_4, leukotriene D_4, and leukotriene E_4 have the ability to contract the bronchial smooth muscles and cause eosinophil locomotion, mucous production, edema, and increased vascular permeability (124). In addition, leukotriene D_4 increases P-selectin-dependent leukocyte rolling flux and eosinophil adhesion through β_2-integrin expression in eosinophils (125, 126). The production and the physiological activity of leukotrienes are inhibited by blocking their receptors or specific enzymes. Cysteinyl leukotriene 1 receptor antagonist (montelukast) is a useful drug for treating allergic rhinitis. However, montelukast is less effective than intranasal corticosteroids.

Mast cells are an important therapeutic target in the treatment of allergic rhinitis. In sensitized patients, Fc receptors and specific IgE antibodies in mast cells can rapidly recognize the previously met allergen, and activated mast cells synthesize and degranulate mediators such as histamine, tryptase, leukotrienes, proteases, cytokines (IL-1, IL-3, IL-4, IL-5, IL-13, TNF-α), and chemokines (IL-8, GM-CSF, MCP-1, RANTES). Intranasal cromolyns are able to inhibit the degranulation of mast cells and the migration or survival of eosinophils resulting in the inhibition of both early and late phases of allergic reactions. Cromolyns can prevent as well as treat the symptoms of allergic rhinitis (127–129).

A large body of evidence regarding the pathways and factors involved in allergic rhinitis suggests possible therapeutic agents for novel management of allergic rhinitis. These factors, which have been extensively examined, include glucocorticoid receptor agonists, histamine receptor antagonists, prostaglandin D_2 receptor antagonists (CRTH2 antagonists), phosphodiesterase 4 inhibitors, bradykinin B_2 receptor antagonists, MAP kinase inhibitors, human neutrophil elastase inhibitors, cytokine synthesis inhibitors, $\alpha_4\beta_1$ integrin antagonists, tryptase inhibitors, ion channel blockers, nitric oxide synthase inhibitors, TLR signaling mediators, and a member of the CD2 subset of the immunoglobulin superfamily receptors (CD244, also known as natural killer cell receptor 2B4) blocker (2, 130). For example, prostaglandin D_2 is one of the major cellular regulators synthesized from arachidonic acid and plays an important role in development and maintenance of allergic response including recruitment of eosinophils (131, 132). CRTH2 receptor antagonists are promising agents for the treatment of chronic allergic diseases including allergic rhinitis (133, 134). Several orally active CRTH2 antagonists are under clinical development (135).

Therapeutic targets related to eosinophils in chronic rhinosinusitis

Chronic rhinosinusitis is usually divided into three subtypes: CRSsNP, CRSwNP, and AFRS (136). The cytokine profile of CRSsNP is mainly Th1 dominant, and the inflammatory cell infiltration is made up of mostly neutrophils, with a low percentage of eosinophils, mast cells, and plasma cells. In contrast, patients with CRSwNP have a predominantly Th2-biased eosinophilic inflammation, and AFRS is defined as chronic rhinosinusitis accompanied by allergic mucin containing degranulated eosinophils and fungal hyphae, and IgE-mediated fungal allergy (3, 6, 7). CRSwNP is associated with increased levels of IL-4, IL-5, IL-13, RANTES, CXCL8, ECP, eotaxin, GM-CSF, VEGF, IgE, ICAM-1, VCAM-1, E-selectin, P-selectin, matrix metalloproteinase (MMP)-1, MMP-2, MMP-7, MMP-9, and Th2-associated transcription factor (GATA-3). These factors play a significant role in the 1) development and activation of eosinophils, 2) downregulation of eosinophilic apoptosis, and 3) eosinophil infiltration in sinonasal sinus mucosa (8, 9, 14, 137–145). CRSwNP is more likely than CRSsNP to be associated with asthma and aspirin-exacerbated lower respiratory disease (7). A close relationship between CRSwNP and Th2-biased eosinophilic inflammation has been reported. However, because CRSwNP sometimes shows neutrophil-dominant inflammation, an analysis of the specific inflammatory pattern (neutrophilic or eosinophilic) may be necessary to adequately treat patients with CRSwNP (146).

Medical treatments for chronic rhinosinusitis include nasal saline irrigation, topical steroid nasal sprays (intranasal glucocorticoids), systemic antibiotics, systemic glucocorticoids, topical steroid irrigations, long-term macrolide treatment, topical antibiotic treatment, antileukotriene treatment, and aspirin desensitization therapy (3). A European position paper on rhinosinusitis and nasal polyps 2012 (EPOS 2012) recommends intranasal glucocorticoids, nasal saline douche, and bacterial lysates for CRSsNP and intranasal glucocorticoids and systemic glucocorticoids for CRSwNP (6). Glucocorticoids and antileukotriene have a significant effect on eosinophilic inflammation in patients with chronic rhinosinusitis, and antileukotriene agents may be used as an adjunct to topical glucocorticoids in the treatment of CRSwNP (147–149).

Monoclonal antibody therapy is one of the immunotherapies using monoclonal antibodies to specifically bind to target cells or proteins. Omalizumab, a humanized

Citation: Advances in Cellular and Molecular Otolaryngology 2015, 3: 26601 - http://dx.doi.org/10.3402/acmo.v3.26601

recombinant monoclonal anti-IgE antibody, reduces circulating levels of IgE, FcεRI expression on mast cells, respiratory tissue eosinophilia, and production of GM-CSF and Th2 cytokines (IL-4, IL-5, and IL-13) (150). A meta-analysis shows that omalizumab is associated with statistically significant symptom relief, decreased rescue medication use, and improvement of quality of life in patients with inadequately controlled allergic chronic rhinosinusitis (151). In addition, a recent double-blind placebo control study showed that mepolizumab, a humanized monoclonal anti-IL-5 antibody, reduces the size of nasal polyps (152). IgE and/or IL-5 inhibition may be considered as a potential novel therapeutic approach in patients with chronic rhinosinusitis with severe eosinophilic inflammation (153). However, EPOS 2012 does not recommend monoclonal antibody therapy for patients with chronic rhinosinusitis (6). The safety and efficacy of monoclonal antibody therapy for chronic rhinosinusitis is under investigation.

There is much debate regarding the role of fungi in chronic rhinosinusitis. Although a significant role of fungi in sinonasal sinus in the majority of chronic rhinosinusitis cases has been suggested, prospective, double-blind, placebo-controlled multicenter clinical trials show that topical amphotericin B treatment has no significant effect on the level of inflammatory mediators and outcome of CRSsNP and CRSwNP (154–156). AFRS, as well as CRSsNP and CRSwNP, is an established subtype of chronic rhinosinusitis, and the diagnostic criteria consist of the following: 1) nasal polyposis, 2) fungi on staining, 3) eosinophilic mucin without fungal invasion into sinus tissue, 4) type I hypersensitivity to fungi, and 5) characteristic radiological findings with soft tissue differential densities on CT scanning (6). Eosinophils play a central role in the onset and development of AFRS. Oral and topical steroids following endoscopic sinus surgery are recommended to control eosinophilic inflammation in AFRS, and the usefulness of antifungal immunotherapy for AFRS has been reported (6, 157).

Therapeutic targets related to eosinophils in asthma

Asthma is a complex and heterogeneous disease with several clinical subtypes. It is often detected in patients with upper respiratory diseases including allergic rhinitis and chronic rhinosinusitis (158, 159). Because an increase of eosinophils in the respiratory tissues and peripheral blood and the overexpression of eosinophil-related inflammatory factors are characteristic findings of most asthma phenotypes, the eosinophil is considered to be the central effector cell responsible for airway inflammation in asthma. Eosinophils have the potential to injure airway tissues through the release of granule-associated basic proteins (which damage nerves and epithelial cells), lipid mediators (which cause bronchoconstriction and mucus hypersecretion), and reactive oxygen species (which generally injure mucosal cells) (93). Serum ECP levels can be used as a clinical tool for estimating eosinophil inflammatory activity in asthma and are also related to disease severity (78).

Inhaled corticosteroids are the recommended drug for patients with asthma. The first choice as add-on therapy to inhaled corticosteroids is an inhaled long-acting β_2 agonist (160). Inhaled corticosteroids strongly inhibit Th2 cytokine release and Th2-driven eosinophilic airway inflammation (161). Inhaled β-agonists are effective at reversing bronchoconstriction in asthma by protein kinase A-dependent relaxation of airway smooth muscle (162). The appropriate combination of long-acting β_2 agonist and inhaled corticosteroid medications is still an important issue, and a recnet cohort study shows that asthma exacerbation is lower for budesonide–formoterol combination therapy versus fluticasone–salmeterol combination therapy due to lower rates of oral corticosteroid use and asthma-related emergency department visits, which indicate better treatment effectiveness of those patients initiated with budesonide–formoterol combination therapy compared with fluticasone–salmeterol combination therapy (163). Because the clinical response to inhaled corticosteroids is variable, the biomarkers in asthmatic patients predicting clinical responsiveness to inhaled corticosteroids therapy are intensively examined (164).

The potential agents in the treatments for difficult asthma include anti-IgE antibody, anti-IL-4 antibody, anti-IL-5 antibody, anti-IL-13 antibody, anti-IL-4 receptor antibody, anti-CXCR2 antibody, and leukotriene receptor blockers (165–167). Omalizumab therapy is an alternative for patients with more severe poorly controlled asthma (168). Mepolizumab significantly reduces asthma exacerbations and is associated with improvements in markers of asthma control (169). A recent meta-analysis shows that the addition of tiotropium may be beneficial for patients with poorly controlled asthma (170). In addition, novel targets in the treatment for asthma such as tyrosine kinase inhibitors, IFN-α, ICAM-1, VCAM-1, integrin, eotaxin/CCR3 pathway, CRTH2, and immune inhibitory receptors on eosinophils [sialic acid-binding immunoglobulin-like lectin (Siglec)-3, Siglec-7, Siglec-8, Siglec-10, FcγRII, CD85a, and CD300a] are intensively examined for future therapeutic uses (4, 32, 43, 48, 93, 113, 171, 172).

Conclusion

Eosinophils are the most important cells involved in the pathophysiology of allergic rhinitis, chronic rhinosinusitis, and asthma. It is also likely that eosinophils play an important role in airway remodeling and inflammation through TGF-β and other inflammatory mediators. Corticosteroids are the most potent and widely used drugs to control eosinophilic inflammation in both the

upper and lower airways. However, steroid-resistant eosinophilic inflammation in the airway has also been reported (122, 173). Several promising agents are under development to increase the number of therapeutic options against eosinophil-associated inflammation. Further studies are needed to characterize molecular mechanisms underlying eosinophilic airway inflammation in each subtype of the diseases.

Acknowledgements

This work was supported by JSPS KAKENHI (Grant-in-Aid for Scientific Research) (Grant Number 25462642).

Conflict of interest and funding

The authors have no conflict of interest to disclose.

References

1. Greiner AN, Hellings PW, Rotiroti G, Scadding GK. Allergic rhinitis. Lancet. 2011; 378: 2112–22. doi: 10.1016/S0140-6736(11)60130-X.
2. Mandhane SN, Shah JH, Thennati R. Allergic rhinitis: an update on disease, present treatments and future prospects. Int Immunopharmacol. 2011; 11: 1646–62. doi: 10.1016/j.intimp.2011.07.005.
3. Hamilos DL. Chronic rhinosinusitis: epidemiology and medical management. J Allergy Clin Immunol. 2011; 128: 693–707; quiz 708–9. doi: 10.1016/j.jaci.2011.08.004.
4. Nakagome K, Nagata M. Pathogenesis of airway inflammation in bronchial asthma. Auris Nasus Larynx. 2011; 38: 555–63. doi: 10.1016/j.anl.2011.01.011.
5. Uhm TG, Kim BS, Chung IY. Eosinophil development, regulation of eosinophil-specific genes, and role of eosinophils in the pathogenesis of asthma. Allergy Asthma Immunol Res. 2012; 4: 68–79. doi: 10.4168/aair.2012.4.2.68.
6. Fokkens WJ, Lund VJ, Mullol J, Bachert C, Alobid I, Baroody F, et al. EPOS 2012: European position paper on rhinosinusitis and nasal polyps 2012. A summary for otorhinolaryngologists. Rhinology. 2012; 50: 1–12. doi: 10.4193/Rhino50E2.
7. Dykewicz MS, Hamilos DL. Rhinitis and sinusitis. J Allergy Clin Immunol. 2010; 125(2 Suppl 2): S103–15. doi: 10.1016/j.jaci.2009.12.989.
8. Van Zele T, Claeys S, Gevaert P, Van Maele G, Holtappels G, Van Cauwenberge P, et al. Differentiation of chronic sinus diseases by measurement of inflammatory mediators. Allergy. 2006; 61: 1280–9. doi: 10.1111/j.1398-9995.2006.01225.x.
9. Bachert C, Gevaert P, Holtappels G, Cuvelier C, van Cauwenberge P. Nasal polyposis: from cytokines to growth. Am J Rhinol. 2000; 14: 279–90. doi: 10.2500/105065800781329573.
10. Robinson S, Douglas R, Wormald PJ. The relationship between atopy and chronic rhinosinusitis. Am J Rhinol. 2006; 20: 625–8. doi: 10.2500/ajr.2006.20.2907.
11. Pawankar R. Nasal polyposis: an update: editorial review. Curr Opin Allergy Clin Immunol. 2003; 3: 1–6.
12. Bateman ND, Fahy C, Woolford TJ. Nasal polyps: still more questions than answers. J Laryngol Otol. 2003; 117: 1–9. doi: 10.1258/002221503321046577.
13. Fokkens W, Lund V, Bachert C, Clement P, Helllings P, Holmstrom M, et al. EAACI position paper on rhinosinusitis and nasal polyps executive summary. Allergy. 2005; 60: 583–601. doi: 10.1111/j.1398-9995.2005.00830.x.
14. Polzehl D, Moeller P, Riechelmann H, Perner S. Distinct features of chronic rhinosinusitis with and without nasal polyps. Allergy. 2006; 61: 1275–9. doi: 10.1111/j.1398-9995.2006.01132.x.
15. Lin H, Lin D, Xiong XS, Dai XX, Lin T. Role of platelet-derived growth factor-a in eosinophilic and non-eosinophilic chronic rhinosinusitis with nasal polyps. Int Forum Allergy Rhinol. 2014; 4: 909–14. doi: 10.1002/alr.21419.
16. Li P, Turner JH. Chronic rhinosinusitis without nasal polyps is associated with increased expression of trefoil factor family peptides. Int Forum Allergy Rhinol. 2014; 4: 571–6. doi: 10.1002/alr.21334.
17. Schlosser RJ. The pathophysiology of chronic rhinosinusitis. Int Forum Allergy Rhinol. 2014; 4: 169–70. doi: 10.1002/alr.21317.
18. Lam M, Hull L, McLachlan R, Snidvongs K, Chin D, Pratt E, et al. Clinical severity and epithelial endotypes in chronic rhinosinusitis. Int Forum Allergy Rhinol. 2013; 3: 121–8. doi: 10.1002/alr.21082.
19. Cho DY, Nayak JV, Bravo DT, Le W, Nguyen A, Edward JA, et al. Expression of dual oxidases and secreted cytokines in chronic rhinosinusitis. Int Forum Allergy Rhinol. 2013; 3: 376–83. doi: 10.1002/alr.21133.
20. Gan EC, Thamboo A, Rudmik L, Hwang PH, Ferguson BJ, Javer AR. Medical management of allergic fungal rhinosinusitis following endoscopic sinus surgery: an evidence-based review and recommendations. Int Forum Allergy Rhinol. 2014; 4: 702–15. doi: 10.1002/alr.21352.
21. Ryan MW. Allergic fungal rhinosinusitis. Otolaryngol Clin North Am. 2011; 44: 697–710, ix–x. doi: 10.1016/j.otc.2011.03.015.
22. Pant H, Schembri MA, Wormald PJ, Macardle PJ. IgE-mediated fungal allergy in allergic fungal sinusitis. Laryngoscope. 2009; 119: 1046–52. doi: 10.1002/lary.20170.
23. Holgate ST. Asthma: a simple concept but in reality a complex disease. Eur J Clin Invest. 2011; 41: 1339–52. doi: 10.1111/j.1365-2362.2011.02534.x.
24. Provost V, Larose MC, Langlois A, Rola-Pleszczynski M, Flamand N, Laviolette M. CCL26/eotaxin-3 is more effective to induce the migration of eosinophils of asthmatics than CCL11/eotaxin-1 and CCL24/eotaxin-2. J Leukoc Biol. 2013; 94: 213–22. doi: 10.1189/jlb.0212074.
25. Lee SY, Yoon SH, Song WJ, Lee SH, Kang HR, Kim SS, et al. Influence of chronic sinusitis and nasal polyp on the lower airway of subjects without lower airway diseases. Allergy Asthma Immunol Res. 2014; 6: 310–15. doi: 10.4168/aair.2014.6.4.310.
26. Brożek JL, Bousquet J, Baena-Cagnani CE, Bonini S, Canonica GW, Casale TB, et al. Allergic rhinitis and its impact on asthma (ARIA) guidelines: 2010 revision. J Allergy Clin Immunol. 2010; 126: 466–76. doi: 10.1016/j.jaci.2010.06.047.
27. Kariyawasam HH, Rotiroti G. Allergic rhinitis, chronic rhinosinusitis and asthma: unravelling a complex relationship. Curr Opin Otolaryngol Head Neck Surg. 2013; 21: 79–86. doi: 10.1097/MOO.0b013e32835ac640.
28. Frieri M. Asthma linked with rhinosinusitis: an extensive review. Allergy Rhinol. 2014; 5: e41–9. doi: 10.2500/ar.2014.5.0083.
29. Lehrer E, Mullol J, Agredo F, Alobid I. Management of chronic rhinosinusitis in asthma patients: is there still a debate? Curr Allergy Asthma Rep. 2014; 14: 440. doi: 10.1007/s11882-014-0440-x.

30. Kariya S, Okano M, Oto T, Higaki T, Makihara S, Haruna T, et al. Pulmonary function in patients with chronic rhinosinusitis and allergic rhinitis. J Laryngol Otol. 2014; 128: 255–62. doi: 10.1017/S0022215114000450.
31. Kariya S, Okano M, Higaki T, Noyama Y, Haruna T, Ishihara H, et al. Chronic rhinosinusitis patients have decreased lung function. Int Forum Allergy Rhinol. 2014; 4: 828–33. doi: 10.1002/alr.21370.
32. Blanchard C, Rothenberg ME. Biology of the eosinophil. Adv Immunol. 2009; 101: 81–121. doi: 10.1016/S0065-2776(08)01003-1.
33. Kita H. Eosinophils: multifunctional and distinctive properties. Int Arch Allergy Immunol. 2013; 161(Suppl 2): 3–9. doi: 10.1159/000350662.
34. Yu C, Cantor AB, Yang H, Browne C, Wells RA, Fujiwara Y, et al. 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. 2002; 195: 1387–95. doi: 10.1084/jem.20020656.
35. Hirasawa R, Shimizu R, Takahashi S, Osawa M, Takayanagi S, Kato Y, et al. Essential and instructive roles of GATA factors in eosinophil development. J Exp Med. 2002; 195: 1379–86. doi: 10.1084/jem.20020170.
36. Iwasaki H, Mizuno S, Mayfield R, Shigematsu H, Arinobu Y, Seed B, et al. Identification of eosinophil lineage-committed progenitors in the murine bone marrow. J Exp Med. 2005; 201: 1891–7. doi: 10.1084/jem.20050548.
37. Wong TW, Jelinek DF. Purification of functional eosinophils from human bone marrow. J Immunol Methods. 2013; 387: 130–9. doi: 10.1016/j.jim.2012.10.006.
38. Kita H. Eosinophils: multifaceted biological properties and roles in health and disease. Immunol Rev. 2011; 242: 161–77. doi: 10.1111/j.1600-065X.2011.01026.x.
39. Mishra A, Hogan SP, Lee JJ, Foster PS, Rothenberg ME. Fundamental signals that regulate eosinophil homing to the gastrointestinal tract. J Clin Invest. 1999; 103: 1719–27. doi: 10.1172/JCI6560.
40. Humbles AA, Lu B, Friend DS, Okinaga S, Lora J, Al-Garawi A, et al. The murine CCR3 receptor regulates both the role of eosinophils and mast cells in allergen-induced airway inflammation and hyperresponsiveness. Proc Natl Acad Sci USA. 2002; 99: 1479–84. doi: 10.1073/pnas.261462598.
41. Pope SM, Fulkerson PC, Blanchard C, Akei HS, Nikolaidis NM, Zimmermann N, et al. Identification of a cooperative mechanism involving interleukin-13 and eotaxin-2 in experimental allergic lung inflammation. J Biol Chem. 2005; 280: 13952–61. doi: 10.1074/jbc.M406037200.
42. Gouon-Evans V, Rothenberg ME, Pollard JW. Postnatal mammary gland development requires macrophages and eosinophils. Development. 2000; 127: 2269–82.
43. Rothenberg ME, Hogan SP. The eosinophil. Annu Rev Immunol. 2006; 24: 147–74. doi: 10.1146/annurev.immunol.24.021605.090720.
44. Rothenberg ME, Owen WF, Jr, Silberstein DS, Soberman RJ, Austen KF, Stevens RL. Eosinophils cocultured with endothelial cells have increased survival and functional properties. Science. 1987; 237: 645–7. doi: 10.1126/science.3110954.
45. Sher A, Coffman RL, Hieny S, Cheever AW. Ablation of eosinophil and IgE responses with anti-IL-5 or anti-IL-4 antibodies fails to affect immunity against *Schistosoma mansoni* in the mouse. J Immunol. 1990; 145: 3911–6.
46. Horie S, Okubo Y, Hossain M, Sato E, Nomura H, Koyama S, et al. Interleukin-13 but not interleukin-4 prolongs eosinophil survival and induces eosinophil chemotaxis. Intern Med. 1997; 36: 179–85. doi: 10.2169/internalmedicine.36.179.
47. Bochner BS, Schleimer RP. The role of adhesion molecules in human eosinophil and basophil recruitment. J Allergy Clin Immunol. 1994; 94: 427–38; quiz 439. doi: 10.1016/0091-6749(94)90195-3.
48. Zimmermann N, Hershey GK, Foster PS, Rothenberg ME. Chemokines in asthma: cooperative interaction between chemokines and IL-13. J Allergy Clin Immunol. 2003; 111: 227–42; quiz 243. doi: 10.1067/mai.2003.139.
49. Zhu Z, Zheng T, Homer RJ, Kim YK, Chen NY, Cohn L, et al. Acidic mammalian chitinase in asthmatic Th2 inflammation and IL-13 pathway activation. Science. 2004; 304: 1678–82. doi: 10.1126/science.1095336.
50. Zeddou M, Delvenne P, El-Shazly AE. Dynamics and function of eosinophils' CD48 molecules in allergic rhinitis and in response to eotaxin stimulation. Adv Cell Mol Otolaryngol. 2013; 1: 22389. doi: 10.3402/acmo.v1i0.22389.
51. Fulkerson PC, Fischetti CA, McBride ML, Hassman LM, Hogan SP, Rothenberg ME. A central regulatory role for eosinophils and the eotaxin/CCR3 axis in chronic experimental allergic airway inflammation. Proc Natl Acad Sci USA. 2006; 103: 16418–23. doi: 10.1073/pnas.0607863103.
52. Pope SM, Zimmermann N, Stringer KF, Karow ML, Rothenberg ME. The eotaxin chemokines and CCR3 are fundamental regulators of allergen-induced pulmonary eosinophilia. J Immunol. 2005; 175: 5341–50. doi: 10.4049/jimmunol.175.8.5341.
53. Nussbaum JC, Van Dyken SJ, von Moltke J, Cheng LE, Mohapatra A, Molofsky AB, et al. Type 2 innate lymphoid cells control eosinophil homeostasis. Nature. 2013; 502: 245–8. doi: 10.1038/nature12526.
54. Kita H, Weiler DA, Abu-Ghazaleh R, Sanderson CJ, Gleich GJ. Release of granule proteins from eosinophils cultured with IL-5. J Immunol. 1992; 149: 629–35.
55. Bracke M, Dubois GR, Bolt K, Bruijnzeel PL, Vaerman JP, Lammers JW, et al. Differential effects of the T helper cell type 2-derived cytokines IL-4 and IL-5 on ligand binding to IgG and IgA receptors expressed by human eosinophils. J Immunol. 1997; 159: 1459–65.
56. Valerius T, Repp R, Kalden JR, Platzer E. Effects of IFN on human eosinophils in comparison with other cytokines. A novel class of eosinophil activators with delayed onset of action. J Immunol. 1990; 145: 2950–8.
57. Saenz SA, Taylor BC, Artis D. Welcome to the neighborhood: epithelial cell-derived cytokines license innate and adaptive immune responses at mucosal sites. Immunol Rev. 2008; 226: 172–90. doi: 10.1111/j.1600-065X.2008.00713.x.
58. Wong CK, Hu S, Cheung PF, Lam CW. Thymic stromal lymphopoietin induces chemotactic and prosurvival effects in eosinophils: implications in allergic inflammation. Am J Respir Cell Mol Biol. 2010; 43: 305–15. doi: 10.1165/rcmb.2009-0168OC.
59. Cherry WB, Yoon J, Bartemes KR, Iijima K, Kita H. A novel IL-1 family cytokine, IL-33, potently activates human eosinophils. J Allergy Clin Immunol. 2008; 121: 1484–90. doi: 10.1016/j.jaci.2008.04.005.
60. Kvarnhammar AM, Cardell LO. Pattern-recognition receptors in human eosinophils. Immunology. 2012; 136: 11–20. doi: 10.1111/j.1365-2567.2012.03556.x.
61. Kvarnhammar AM, Petterson T, Cardell LO. NOD-like receptors and RIG-I-like receptors in human eosinophils: activation by NOD1 and NOD2 agonists. Immunology. 2011; 134: 314–25. doi: 10.1111/j.1365-2567.2011.03492.x.
62. Wong CK, Cheung PF, Ip WK, Lam CW. Intracellular signaling mechanisms regulating toll-like receptor-mediated

activation of eosinophils. Am J Respir Cell Mol Biol. 2007; 37: 85–96. doi: 10.1165/rcmb.2006-0457OC.
63. Plötz SG, Lentschat A, Behrendt H, Plötz W, Hamann L, Ring J, et al. The interaction of human peripheral blood eosinophils with bacterial lipopolysaccharide is CD14 dependent. Blood. 2001; 97: 235–41. doi: 10.1182/blood.V97.1.235.
64. Sabroe I, Jones EC, Usher LR, Whyte MK, Dower SK. Toll-like receptor (TLR)2 and TLR4 in human peripheral blood granulocytes: a critical role for monocytes in leukocyte lipopolysaccharide responses. J Immunol. 2002; 168: 4701–10. doi: 10.4049/jimmunol.168.9.4701.
65. Nagase H, Okugawa S, Ota Y, Yamaguchi M, Tomizawa H, Matsushima K, et al. Expression and function of Toll-like receptors in eosinophils: activation by Toll-like receptor 7 ligand. J Immunol. 2003; 171: 3977–82. doi: 10.4049/jimmunol.171.8.3977.
66. Månsson A, Fransson M, Adner M, Benson M, Uddman R, Björnsson S, et al. TLR3 in human eosinophils: functional effects and decreased expression during allergic rhinitis. Int Arch Allergy Immunol. 2010; 151: 118–28. doi: 10.1159/000236001.
67. Janke M, Poth J, Wimmenauer V, Giese T, Coch C, Barchet W, et al. Selective and direct activation of human neutrophils but not eosinophils by Toll-like receptor 8. J Allergy Clin Immunol. 2009; 123: 1026–33. doi: 10.1016/j.jaci.2009.02.015.
68. Driss V, Legrand F, Hermann E, Loiseau S, Guerardel Y, Kremer L, et al. TLR2-dependent eosinophil interactions with mycobacteria: role of alpha-defensins. Blood. 2009; 113: 3235–44. doi: 10.1182/blood-2008-07-166595.
69. Miike S, McWilliam AS, Kita H. Trypsin induces activation and inflammatory mediator release from human eosinophils through protease-activated receptor-2. J Immunol. 2001; 167: 6615–22. doi: 10.4049/jimmunol.167.11.6615.
70. Miike S, Kita H. Human eosinophils are activated by cysteine proteases and release inflammatory mediators. J Allergy Clin Immunol. 2003; 111: 704–13. doi: 10.1067/mai.2003.1332.
71. Matsuwaki Y, Wada K, White TA, Benson LM, Charlesworth MC, Checkel JL, et al. Recognition of fungal protease activities induces cellular activation and eosinophil-derived neurotoxin release in human eosinophils. J Immunol. 2009; 183: 6708–16. doi: 10.4049/jimmunol.0901220.
72. Wada K, Matsuwaki Y, Yoon J, Benson LM, Checkel JL, Bingemann TA, et al. Inflammatory responses of human eosinophils to cockroach are mediated through protease-dependent pathways. J Allergy Clin Immunol. 2010; 126: 169–72.e2. doi: 10.1016/j.jaci.2010.04.007.
73. Bush RK, Prochnau JJ. Alternaria-induced asthma. J Allergy Clin Immunol. 2004; 113: 227–34. doi: 10.1016/j.jaci.2003.11.023.
74. Bianchi ME. DAMPs, PAMPs and alarmins: all we need to know about danger. J Leukoc Biol. 2007; 81: 1–5. doi: 10.1189/jlb.0306164.
75. Kobayashi T, Kouzaki H, Kita H. Human eosinophils recognize endogenous danger signal crystalline uric acid and produce proinflammatory cytokines mediated by autocrine ATP. J Immunol. 2010; 184: 6350–8. doi: 10.4049/jimmunol.0902673.
76. Lotfi R, Herzog GI, DeMarco RA, Beer-Stolz D, Lee JJ, Rubartelli A, et al. Eosinophils oxidize damage-associated molecular pattern molecules derived from stressed cells. J Immunol. 2009; 183: 5023–31. doi: 10.4049/jimmunol.0900504.
77. Malik A, Batra JK. Antimicrobial activity of human eosinophil granule proteins: involvement in host defence against pathogens. Crit Rev Microbiol. 2012; 38: 168–81. doi: 10.3109/1040841X.2011.645519.
78. Acharya KR, Ackerman SJ. Eosinophil granule proteins: form and function. J Biol Chem. 2014; 289: 17406–15. doi: 10.1074/jbc.R113.546218.
79. Plager DA, Loegering DA, Checkel JL, Tang J, Kephart GM, Caffes PL, et al. Major basic protein homolog (MBP2): a specific human eosinophil marker. J Immunol. 2006; 177: 7340–5. doi: 10.4049/jimmunol.177.10.7340.
80. Barker RL, Loegering DA, Ten RM, Hamann KJ, Pease LR, Gleich GJ. Eosinophil cationic protein cDNA. Comparison with other toxic cationic proteins and ribonucleases. J Immunol. 1989; 143: 952–5.
81. Rosenberg HF. Recombinant human eosinophil cationic protein. Ribonuclease activity is not essential for cytotoxicity. J Biol Chem. 1995; 270: 7876–81. doi: 10.1074/jbc.270.14.7876.
82. Domachowske JB, Dyer KD, Adams AG, Leto TL Rosenberg HF. Eosinophil cationic protein/RNase 3 is another RNase A-family ribonuclease with direct antiviral activity. Nucleic Acids Res. 1998; 26: 3358–63. doi: 10.1093/nar/26.14.3358.
83. Torrent M, Pulido D, Nogués MV, Boix E. Exploring new biological functions of amyloids: bacteria cell agglutination mediated by host protein aggregation. PLoS Pathog. 2012; 8: e1003005. doi: 10.1371/journal.ppat.1003005.
84. Pulido D, Torrent M, Andreu D, Nogués MV, Boix E. Two human host defense ribonucleases against mycobacteria, the eosinophil cationic protein (RNase 3) and RNase 7. Antimicrob Agents Chemother. 2013; 57: 3797–805. doi: 10.1128/AAC.00428-13.
85. Abu-Ghazaleh RI, Dunnette SL, Loegering DA, Checkel JL, Kita H, Thomas LL, et al. Eosinophil granule proteins in peripheral blood granulocytes. J Leukoc Biol. 1992; 52: 611–8.
86. Tang XN, Zheng Z, Giffard RG, Yenari MA. Significance of marrow-derived nicotinamide adenine dinucleotide phosphate oxidase in experimental ischemic stroke. Ann Neurol. 2011; 70: 606–15. doi: 10.1002/ana.22476.
87. Comhair SA, Erzurum SC. Redox control of asthma: molecular mechanisms and therapeutic opportunities. Antioxid Redox Signal. 2010; 12: 93–124. doi: 10.1089/ARS.2008.2425.
88. Auriault C, Capron M, Capron A. Activation of rat and human eosinophils by soluble factor(s) released by *Schistosoma mansoni* schistosomula. Cell Immunol. 1982; 66: 59–69. doi: 10.1016/0008-8749(82)90157-5.
89. Locksley RM, Wilson CB, Klebanoff SJ. Role for endogenous and acquired peroxidase in the toxoplasmacidal activity of murine and human mononuclear phagocytes. J Clin Invest. 1982; 69: 1099–111. doi: 10.1172/JCI110545.
90. Henderson WR, Jörg A, Klebanoff SJ. Eosinophil peroxidase-mediated inactivation of leukotrienes B4, C4, and D4. J Immunol. 1982; 128: 2609–13.
91. Henderson WR, Jong EC, Klebanoff SJ. Binding of eosinophil peroxidase to mast cell granules with retention of peroxidatic activity. J Immunol. 1980; 124: 1383–8.
92. Boix E, Nogués MV. Mammalian antimicrobial proteins and peptides: overview on the RNase A superfamily members involved in innate host defence. Mol Biosyst. 2007; 3: 317–35. doi: 10.1039/B617527A.
93. Hogan SP, Rosenberg HF, Moqbel R, Phipps S, Foster PS, Lacy P, et al. Eosinophils: biological properties and role in health and disease. Clin Exp Allergy. 2008; 38: 709–50. doi: 10.1111/j.1365-2222.2008.02958.x.
94. Harrison AM, Bonville CA, Rosenberg HF, Domachowske JB. Respiratory syncytical virus-induced chemokine expression in the lower airways: eosinophil recruitment and degranulation.

Am J Respir Crit Care Med. 1999; 159: 1918–24. doi: 10.1164/ajrccm.159.6.9805083.
95. Domachowske JB, Dyer KD, Bonville CA, Rosenberg HF. Recombinant human eosinophil-derived neurotoxin/RNase 2 functions as an effective antiviral agent against respiratory syncytial virus. J Infect Dis. 1998; 177: 1458–64. doi: 10.1086/515322.
96. Yang D, Chen Q, Rosenberg HF, Rybak SM, Newton DL, Wang ZY, et al. Human ribonuclease A superfamily members, eosinophil-derived neurotoxin and pancreatic ribonuclease, induce dendritic cell maturation and activation. J Immunol. 2004; 173: 6134–42. doi: 10.4049/jimmunol.173.10.6134.
97. Yang D, Chen Q, Su SB, Zhang P, Kurosaka K, Caspi RR, et al. Eosinophil-derived neurotoxin acts as an alarmin to activate the TLR2-MyD88 signal pathway in dendritic cells and enhances Th2 immune responses. J Exp Med. 2008; 205: 79–90. doi: 10.1084/jem.20062027.
98. Kim KS, Won HR, Park CY, Hong JH, Lee JH, Lee KE, et al. Analyzing serum eosinophil cationic protein in the clinical assessment of chronic rhinosinusitis. Am J Rhinol Allergy. 2013; 27: e75–80. doi: 10.2500/ajra.2013.27.3901.
99. Ponikau JU, Winter LA, Kephart GM, Squillace DL, Hershcovitch MD, Moon S, et al. An immunologic test for chronic rhinosinusitis based on free intranasal eosinophilic major basic protein. Int Forum Allergy Rhinol. 2015; 5: 28–35. doi: 10.1002/alr.21421.
100. Jung JW, Kang HR, Lee HS, Park HW, Cho SH, Min KU, et al. Expression levels of eosinophil granule protein mRNAs in induced sputum reflect airway hyperresponsiveness and airflow limitation. Tohoku J Exp Med. 2014; 233: 49–56. doi: 10.1620/tjem.233.49.
101. Bystrom J, Patel SY, Amin K, Bishop-Bailey D. Dissecting the role of eosinophil cationic protein in upper airway disease. Curr Opin Allergy Clin Immunol. 2012; 12: 18–23. doi: 10.1097/ACI.0b013e32834eccaf.
102. Bystrom J, Amin K, Bishop-Bailey D. Analysing the eosinophil cationic protein--a clue to the function of the eosinophil granulocyte. Respir Res. 2011; 12: 10. doi: 10.1186/1465-9921-12-10.
103. Khoury P, Grayson PC, Klion AD. Eosinophils in vasculitis: characteristics and roles in pathogenesis. Nat Rev Rheumatol. 2014; 10: 474–83. doi: 10.1038/nrrheum.2014.98.
104. Shamri R, Xenakis JJ, Spencer LA. Eosinophils in innate immunity: an evolving story. Cell Tissue Res. 2011; 343: 57–83. doi: 10.1007/s00441-010-1049-6.
105. Yamamura K, Adachi T, Masuda T, Kojima Y, Hara A, Toda T, et al. Intracellular protein phosphorylation in eosinophils and the functional relevance in cytokine production. Int Arch Allergy Immunol. 2009; 149(Suppl 1): 45–50. doi: 10.1159/000210653.
106. Curran CS, Bertics PJ. Lactoferrin regulates an axis involving CD11b and CD49d integrins and the chemokines MIP-1α and MCP-1 in GM-CSF-treated human primary eosinophils. J Interferon Cytokine Res. 2012; 32: 450–61. doi: 10.1089/jir.2011.0111.
107. Nutku E, Zhuang Q, Soussi-Gounni A, Aris F, Mazer BD, Hamid Q. Functional expression of IL-12 receptor by human eosinophils: IL-12 promotes eosinophil apoptosis. J Immunol. 2001; 167: 1039–46. doi: 10.4049/jimmunol.167.2.1039.
108. Cirino G, Vergnolle N. Proteinase-activated receptors (PARs): crossroads between innate immunity and coagulation. Curr Opin Pharmacol. 2006; 6: 428–34. doi: 10.1016/j.coph.2006.05.001.
109. Shpacovitch V, Feld M, Bunnett NW, Steinhoff M. Protease-activated receptors: novel PARtners in innate immunity. Trends Immunol. 2007; 28: 541–50. doi: 10.1016/j.it.2007.09.001.
110. Inoue Y, Matsuwaki Y, Shin SH, Ponikau JU, Kita H. Nonpathogenic, environmental fungi induce activation and degranulation of human eosinophils. J Immunol. 2005; 175: 5439–47. doi: 10.4049/jimmunol.175.8.5439.
111. Yoon J, Ponikau JU, Lawrence CB, Kita H. Innate antifungal immunity of human eosinophils mediated by a β2 integrin, CD11b. J Immunol. 2008; 181: 2907–15. doi: 10.4049/jimmunol.181.4.2907.
112. Munitz A, Bachelet I, Fraenkel S, Katz G, Mandelboim O, Simon HU, et al. 2B4 (CD244) is expressed and functional on human eosinophils. J Immunol. 2005; 174: 110–8. doi: 10.4049/jimmunol.174.1.110.
113. Munitz A Levi-Schaffer F. Inhibitory receptors on eosinophils: a direct hit to a possible Achilles heel? J Allergy Clin Immunol. 2007; 119: 1382–7. doi: 10.1016/j.jaci.2007.01.031.
114. Dunne DW, Richardson BA, Jones FM, Clark M, Thorne KJ, Butterworth AE. The use of mouse/human chimaeric antibodies to investigate the roles of different antibody isotypes, including IgA2, in the killing of *Schistosoma mansoni* schistosomula by eosinophils. Parasite Immunol. 1993; 15: 181–5. doi: 10.1111/j.1365-3024.1993.tb00598.x.
115. Jönsson F, Mancardi DA, Zhao W, Kita Y, Iannascoli B, Khun H, et al. Human FcγRIIA induces anaphylactic and allergic reactions. Blood. 2012; 119: 2533–44. doi: 10.1182/blood-2011-07-367334.
116. Watanabe T, Okano M, Hattori H, Yoshino T, Ohno N, Ohta N, et al. Roles of FcγRIIB in nasal eosinophilia and IgE production in murine allergic rhinitis. Am J Respir Crit Care Med. 2004; 169: 105–12. doi: 10.1164/rccm.200302-239OC.
117. Horie S, Kita H. CD11b/CD18 (Mac-1) is required for degranulation of human eosinophils induced by human recombinant granulocyte-macrophage colony-stimulating factor and platelet-activating factor. J Immunol. 1994; 152: 5457–67.
118. Kato M, Abraham RT, Okada S, Kita H. Ligation of the β_2 integrin triggers activation and degranulation of human eosinophils. Am J Respir Cell Mol Biol. 1998; 18: 675–86. doi: 10.1165/ajrcmb.18.5.2885.
119. Nagata M, Sedgwick JB, Kita H, Busse WW. Granulocyte macrophage colony-stimulating factor augments ICAM-1 and VCAM-1 activation of eosinophil function. Am J Respir Cell Mol Biol. 1998; 19: 158–66. doi: 10.1165/ajrcmb.19.1.3001.
120. Gounni AS, Lamkhioued B, Ochiai K, Tanaka Y, Delaporte E, Capron A, et al. High-affinity IgE receptor on eosinophils is involved in defence against parasites. Nature. 1994; 367: 183–6. doi: 10.1038/367183a0.
121. Ciebiada M, Gorska-Ciebiada M, Gorski P. sICAM-1 and TNF-α in asthma and rhinitis: relationship with the presence of atopy. J Asthma. 2011; 48: 660–6. doi: 10.3109/02770903.2011.604886.
122. Barnes PJ. Mechanisms and resistance in glucocorticoid control of inflammation. J Steroid Biochem Mol Biol. 2010; 120: 76–85. doi: 10.1016/j.jsbmb.2010.02.018.
123. Karaki M, Akiyama K, Mori N. Efficacy of intranasal steroid spray (mometasone furoate) on treatment of patients with seasonal allergic rhinitis: comparison with oral corticosteroids. Auris Nasus Larynx. 2013; 40: 277–81. doi: 10.1016/j.anl.2012.09.004.
124. Fregonese L, Silvestri M, Sabatini F, Rossi GA. Cysteinyl leukotrienes induce human eosinophil locomotion and adhesion molecule expression via a $CysLT_1$ receptor-mediated mechanism. Clin Exp Allergy. 2002; 32: 745–50. doi: 10.1046/j.1365-2222.2002.01384.x.
125. Busse W, Kraft M. Cysteinyl leukotrienes in allergic inflammation: strategic target for therapy. Chest. 2005; 127: 1312–26. doi: 10.1378/chest.127.4.1312.

126. Nagata M, Saito K, Tsuchiya K, Sakamoto Y. Leukotriene D_4 upregulates eosinophil adhesion via the cysteinyl leukotriene 1 receptor. J Allergy Clin Immunol. 2002; 109: 676–80. doi: 10.1067/mai.2002.122841.
127. Gschwentner M, Susanna A, Schmarda A, Laich A, Nagl UO, Ellemunter H, et al. I_{Cln}: a chloride channel paramount for cell volume regulation. J Allergy Clin Immunol. 1996; 98(5 Pt 2): S98–101; discussion: S105–6. doi: 10.1016/S0091-6749(96)70023-4.
128. Stenton GR, Chow SM, Lau HY. Inhibition of rat peritoneal mast cell exocytosis by frusemide: a comparison with disodium cromoglycate. Life Sci. 1998; 62: PL49–54. doi: 10.1016/S0024-3205(97)01107-7.
129. Holgate ST. Reflections on the mechanism(s) of action of sodium cromoglycate (Intal) and the role of mast cells in asthma. Respir Med. 1989; 83(Suppl A): 25–31. doi: 10.1016/S0954-6111(89)80247-1.
130. El-Shazly AE, Henket M, Lefebvre PP, Louis R. 2B4 (CD244) is involved in eosinophil adhesion and chemotaxis, and its surface expression is increased in allergic rhinitis after challenge. Int J Immunopathol Pharmacol. 2011; 24: 949–60.
131. Nomiya R, Okano M, Fujiwara T, Maeda M, Kimura Y, Kino K, et al. CRTH2 plays an essential role in the pathophysiology of Cry j 1-induced pollinosis in mice. J Immunol. 2008; 180: 5680–8. doi: 10.4049/jimmunol.180.8.5680.
132. Okano M, Fujiwara T, Sugata Y, Gotoh D, Masaoka Y, Sogo M, et al. Presence and characterization of prostaglandin D2-related molecules in nasal mucosa of patients with allergic rhinitis. Am J Rhinol. 2006; 20: 342–8. doi: 10.2500/ajr.2006.20.2865.
133. El-Shazly AE, Begon DY, Kustermans G, Arafa M, Dortu E, Henket M, et al. Novel association between vasoactive intestinal peptide and CRTH2 receptor in recruiting eosinophils: a possible biochemical mechanism for allergic eosinophilic inflammation of the airways. J Biol Chem. 2013; 288: 1374–84. doi: 10.1074/jbc.M112.422675.
134. El-Shazly AE, Moonen V, Mawet M, Begon D, Henket M, Arafa M, et al. IFN-γ and TNF-α potentiate prostaglandin D2-induced human eosinophil chemotaxis through up-regulation of CRTH2 surface receptor. Int Immunopharmacol. 2011; 11: 1864–70. doi: 10.1016/j.intimp.2011.07.017.
135. Ulven T, Kostenis E. Novel CRTH2 antagonists: a review of patents from 2006 to 2009. Expert Opin Ther Pat. 2010; 20: 1505–30. doi: 10.1517/13543776.2010.525506.
136. Eloy P, Poirrier AL, De Dorlodot C, Van Zele T, Watelet JB, Bertrand B. Actual concepts in rhinosinusitis: a review of clinical presentations, inflammatory pathways, cytokine profiles, remodeling, and management. Curr Allergy Asthma Rep. 2011; 11: 146–62. doi: 10.1007/s11882-011-0180-0.
137. Van Bruaene N, Pérez-Novo CA, Basinski TM, Van Zele T, Holtappels G, De Ruyck N, et al. T-cell regulation in chronic paranasal sinus disease. J Allergy Clin Immunol. 2008; 121: 1435–41, 1441.e1–3. doi: 10.1016/j.jaci.2008.02.018.
138. Li HB, Cai KM, Liu Z, Xia JH, Zhang Y, Xu R, et al. Foxp3+ T regulatory cells (Tregs) are increased in nasal polyps (NP) after treatment with intranasal steroid. Clin Immunol. 2008; 129: 394–400. doi: 10.1016/j.clim.2008.07.031.
139. Pérez-Novo CA, Kowalski ML, Kuna P, Ptasinska A, Holtappels G, van Cauwenberge P, et al. Aspirin sensitivity and IgE antibodies to *Staphylococcus aureus* enterotoxins in nasal polyposis: studies on the relationship. Int Arch Allergy Immunol. 2004; 133: 255–60. doi: 10.1159/000076832.
140. Wise SK, Laury AM, Katz EH, Den Beste KA, Parkos CA, Nusrat A. Interleukin-4 and interleukin-13 compromise the sinonasal epithelial barrier and perturb intercellular junction protein expression. Int Forum Allergy Rhinol. 2014; 4: 361–70. doi: 10.1002/alr.21298.
141. Ebert CS, Jr, McKinney KA, Urrutia G, Wu M, Rose AS, Fleischman GM, et al. Expression of protease-activated receptors in allergic fungal rhinosinusitis. Int Forum Allergy Rhinol. 2014; 4: 266–71. doi: 10.1002/alr.21295.
142. Nonaka M, Pawankar R, Saji F, Yagi T. Eotaxin synthesis by nasal polyp fibroblasts. Acta Otolaryngol. 1999; 119: 816–20. doi: 10.1080/00016489950180478.
143. Nonaka M, Pawankar R, Saji F, Yagi T. Distinct expression of RANTES and GM-CSF by lipopolysaccharide in human nasal fibroblasts but not in other airway fibroblasts. Int Arch Allergy Immunol. 1999; 119: 314–21. doi: 10.1159/000024209.
144. Simon HU, Yousefi S, Schranz C, Schapowal A, Bachert C, Blaser K. Direct demonstration of delayed eosinophil apoptosis as a mechanism causing tissue eosinophilia. J Immunol. 1997; 158: 3902–8.
145. De Corso E, Baroni S, Battista M, Romanello M, Penitente R, Di Nardo W, et al. Nasal fluid release of eotaxin-3 and eotaxin-2 in persistent sinonasal eosinophilic inflammation. Int Forum Allergy Rhinol. 2014; 4: 617–24. doi: 10.1002/alr.21348.
146. Bachert C, Claeys SE, Tomassen P, van Zele T, Zhang N. Rhinosinusitis and asthma: a link for asthma severity. Curr Allergy Asthma Rep. 2010; 10: 194–201. doi: 10.1007/s11882-010-0096-0.
147. Ragab S, Parikh A, Darby YC, Scadding GK. An open audit of montelukast, a leukotriene receptor antagonist, in nasal polyposis associated with asthma. Clin Exp Allergy. 2001 Sep; 31(9): 1385–91. doi: 10.1046/j.1365-2222.2001.01160.x.
148. Stewart RA, Ram B, Hamilton G, Weiner J, Kane KJ. Montelukast as an adjunct to oral and inhaled steroid therapy in chronic nasal polyposis. Otolaryngol Head Neck Surg. 2008; 139: 682–7. doi: 10.1016/j.otohns.2008.07.010.
149. Steinke JW, Kennedy JL. Leukotriene inhibitors in sinusitis. Curr Infect Dis Rep. 2012; 14: 147–54. doi: 10.1007/s11908-012-0245-9.
150. Holgate S, Smith N, Massanari M, Jimenez P. Effects of omalizumab on markers of inflammation in patients with allergic asthma. Allergy. 2009; 64: 1728–36. doi: 10.1111/j.1398-9995.2009.02201.x.
151. Tsabouri S, Tseretopoulou X, Priftis K, Ntzani EE. Omalizumab for the treatment of inadequately controlled allergic rhinitis: a systematic review and meta-analysis of randomized clinical trials. J Allergy Clin Immunol Pract. 2014; 2: 332–40.e1. doi: 10.1016/j.jaip.2014.02.001.
152. Gevaert P, Van Bruaene N, Cattaert T, Van Steen K, Van Zele T, Acke F, et al. Mepolizumab, a humanized anti-IL-5 mAb, as a treatment option for severe nasal polyposis. J Allergy Clin Immunol. 2011; 128: 989–95.e1–8. doi: 10.1016/j.jaci.2011.07.056.
153. Bachert C, Zhang N. Chronic rhinosinusitis and asthma: novel understanding of the role of IgE 'above atopy'. J Intern Med. 2012; 272: 133–43. doi: 10.1111/j.1365-2796.2012.02559.x.
154. Ponikau JU, Sherris DA, Kern EB, Homburger HA, Frigas E, Gaffey TA, et al. The diagnosis and incidence of allergic fungal sinusitis. Mayo Clin Proc. 1999; 74: 877–84. doi: 10.4065/74.9.877.
155. Ebbens FA, Georgalas C, Luiten S, van Drunen CM, Badia L, Scadding GK, et al. The effect of topical amphotericin B on inflammatory markers in patients with chronic rhinosinusitis: a multicenter randomized controlled study. Laryngoscope. 2009; 119: 401–8. doi: 10.1002/lary.20064.
156. Ebbens FA, Scadding GK, Badia L, Hellings PW, Jorissen M, Mullol J, et al. Amphotericin B nasal lavages: not a solution

for patients with chronic rhinosinusitis. J Allergy Clin Immunol. 2006; 118: 1149–56. doi: 10.1016/j.jaci.2006.07.058.
157. Greenhaw B, deShazo RD, Arnold J, Wright L. Fungal immunotherapy in patients with allergic fungal sinusitis. Ann Allergy Asthma Immunol. 2011; 107: 432–6. doi: 10.1016/j.anai.2011.05.021.
158. Reisacher WR. Asthma and the otolaryngologist. Int Forum Allergy Rhinol. 2014; 4(Suppl 2): S70–3. doi: 10.1002/alr.21386.
159. Kariya S, Okano M, Nishizaki K. An association between upper and lower respiratory tract diseases. Ann Otolaryngol Rhinol. 2014; 1: 1001.
160. British Thoracic Society Scottish Intercollegiate Guidelines Network. British guideline on the management of asthma. Thorax. 2014; 69(Suppl 1): i1–192.
161. Brusselle G, Bracke K. Targeting immune pathways for therapy in asthma and chronic obstructive pulmonary disease. Ann Am Thorac Soc. 2014; 11(Suppl 5): S322–8. doi: 10.1513/AnnalsATS.201403-118AW.
162. Morgan SJ, Deshpande DA, Tiegs BC, Misior AM, Yan H, Hershfeld AV, et al. β-Agonist-mediated relaxation of airway smooth muscle is protein kinase A-dependent. J Biol Chem. 2014; 289: 23065–74. doi: 10.1074/jbc.M114.557652.
163. Tunceli O, Williams SA, Kern DM, Elhefni H, Pethick N, Wessman C, et al. Comparative effectiveness of budesonide–formoterol combination and fluticasone–salmeterol combination for asthma management: a United States retrospective database analysis. J Allergy Clin Immunol Pract. 2014; 2: 719–26.e6. doi: 10.1016/j.jaip.2014.07.016.
164. Cowan DC, Taylor DR, Peterson LE, Cowan JO, Palmay R, Williamson A, et al. Biomarker-based asthma phenotypes of corticosteroid response. J Allergy Clin Immunol. 2014 Dec 6. doi: 10.1016/j.jaci.2014.10.026. [in press].
165. Busse WW, Ring J, Huss-Marp J, Kahn JE. A review of treatment with mepolizumab, an anti-IL-5 mAb, in hypereosinophilic syndromes and asthma. J Allergy Clin Immunol. 2010; 125: 803–13. doi: 10.1016/j.jaci.2009.11.048.
166. Molfino NA, Gossage D, Kolbeck R, Parker JM, Geba GP. Molecular and clinical rationale for therapeutic targeting of interleukin-5 and its receptor. Clin Exp Allergy. 2012; 42: 712–37. doi: 10.1111/j.1365-2222.2011.03854.x.
167. Chung KF, Wenzel SE, Brożek JL, Bush A, Castro M, Sterk PJ, et al. International ERS/ATS guidelines on definition, evaluation and treatment of severe asthma. Eur Respir J. 2014; 43: 343–73. doi: 10.1183/09031936.00202013.
168. Hendeles L, Khan YR, Shuster JJ, Chesrown SE, Abu-Hasan M. Omalizumab therapy for asthma patients with poor adherence to inhaled corticosteroid therapy. Ann Allergy Asthma Immunol. 2015; 114: 58–62.e2. doi: 10.1016/j.anai.2014.10.012.
169. Ortega HG, Liu MC, Pavord ID, Brusselle GG, FitzGerald JM, Chetta A, et al. Mepolizumab treatment in patients with severe eosinophilic asthma. N Engl J Med. 2014; 371: 1198–207. doi: 10.1056/NEJMoa1403290.
170. Lee SW, Kim HJ, Yoo KH, Park YB, Park JY, Jung JY, et al. Long-acting anticholinergic agents in patients with uncontrolled asthma: a systematic review and meta-analysis. Int J Tuberc Lung Dis. 2014; 18: 1421–30. doi: 10.5588/ijtld.14.0275.
171. Nutku E, Aizawa H, Hudson SA, Bochner BS. Ligation of Siglec-8: a selective mechanism for induction of human eosinophil apoptosis. Blood. 2003; 101: 5014–20. doi: 10.1182/blood-2002-10-3058.
172. Shik D, Munitz A. Regulation of allergic inflammatory responses by inhibitory receptors. Clin Exp Allergy. 2010; 40: 700–9. doi: 10.1111/j.1365-2222.2010.03501.x.
173. Yang M, Kumar RK, Foster PS. Pathogenesis of steroid-resistant airway hyperresponsiveness: interaction between IFN-γ and TLR4/MyD88 pathways. J Immunol. 2009; 182: 5107–15. doi: 10.4049/jimmunol.0803468.