Review

Multiple Biological Aspects of Eosinophils in Host Defense, Eosinophil-Associated Diseases, Immunoregulation, and Homeostasis: Is Their Role Beneficial, Detrimental, Regulator, or Bystander?

Akira Kanda,*a,b Yun Yasutaka,a Dan Van Bui,a Kensuke Suzuki,a Shunsuke Sawada,a Yoshiki Kobayashi,a,b Mikiya Asako,a,b and Hiroshi Iwaia

*a Department of Otolaryngology, Head and Neck Surgery, Kansai Medical University; 2–5–1 Shinmachi, Hirakata, Osaka 573–1010, Japan; and b Allergy Center, Kansai Medical University; 2–5–1 Shinmachi, Hirakata, Osaka 573–1010, Japan.

Received October 10, 2019

Eosinophils are innate immune leukocytes and play important roles as terminal effector cells owing to their mediators, such as tissue-destructive cationic proteins, cytokines, chemokines, and lipid mediators. Historically, they are not only considered important in host defense against parasitic, viral, fungal, and bacterial infections but also implicated in the pathogenesis of eosinophil-associated diseases, such as allergic rhinitis, asthma, eosinophilic chronic rhinosinusitis, esophagitis, atopic dermatitis, myopathies, and hypereosinophilic syndrome. Moreover, recent studies have shown that eosinophils have an immune regulatory and homeostatic function. Interestingly, there is emerging evidence that eosinophils are accumulated through adoptive T-helper 2 (Th2) and innate Th2 responses, mechanisms of the classical allergen-specific immunoglobulin E (IgE)-mediated response, and group 2 innate lymphoid cell-derived interleukin-5, respectively. Furthermore, in agreement with current concepts of eosinophil subtypes, it has been shown that resident and phenotypically distinct eosinophils, i.e., resident and recruited inflammatory eosinophils, exist in inflamed sites, and each has different functions. Thus, the classical and novel studies suggest that eosinophils have multiple functions, and their roles may be altered by the environment. In this article, we review multiple biological aspects of eosinophils (novel and classical roles), including their beneficial and detrimental effects, immunoregulation, and homeostatic function.

Key words allergic rhinitis (AR); asthma; eosinophil; eosinophilic chronic rhinosinusitis (ECRS)

1. MULTIPLE BIOLOGICAL FUNCTIONS OF EOSINOPHILS

Inflammatory reaction following an inflammatory cascade induced by stimuli (e.g., tissue chemical and physical injury, infection, allergen, allograft, and tumor) is a critical step in host defense. However, persistent inflammation may cause tissue damage with/without dysfunction and is life-threatening in patients with severe asthma. In the case of allergic inflammation (T-helper 2 (Th2) pathology such as allergic rhinitis (AR) and induced asthma), bridging of specific immunoglobulin E (IgE) receptors on mast cells and antigen results in degranulation from mast cells and induces an allergic response. Subsequently, inflammatory cells infiltrate the inflamed tissue through cytokine and chemokine networks. One of the characteristics of allergic reaction is infiltration of eosinophils in the tissue within several hours, and eosinophils play a critical role in the development of this Th2 pathogenesis.1,2

In 1879, Ehrlich first mentioned cells stained by eosin, which is a bright red synthetic dye produced by the action of bromine on fluorescein and stains basic proteins owing to its acidic nature.3 Until the 1970s, eosinophils were thought to play an antiinflammatory role as sweepers in inflamed tissue. These cells contain enzymes exerting a neutralizing effect against proinflammatory mediators and appear in the inflamed site in the late-phase allergic response.1,2,5 Following the discovery of cytotoxic proteins (i.e., major basic protein (MBP), eosinophil peroxidase (EPO), eosinophil cationic protein (ECP), and eosinophil-derived neurotoxin (EDN)) in eosinophils in the 1980s, these cells have also been considered to play a detrimental role as terminal effector cells in allergic inflammation.3 Moreover, it was revealed that eosinophils contain Th2 cytokines (interleukin (IL)-4, IL-5, IL-9, IL-13, and IL-25), Th1 cytokines (IL-12 and interferon (IFN)-γ), acute proinflammatory cytokines (tumor necrosis factor (TNF)-α, IL-1β, IL-6, and IL-8), chemokines (regulated on activation, normal T cell expressed and secreted, and eotaxin), and lipid mediators (platelet-activating factor and leukotriene C4) (Fig. 1).

Numerous reports clarified that eosinophils have specific granule contents and lipid bodies and express a large variety of receptors, such as chemotactant receptors, cytokine and growth factor receptors, adhesion receptors, pattern-recognition receptors, Fc receptors, and receptors for lipid mediators.1,2,5 (Fig. 1). Thus, these multiple eosinophil-derived mediators and large variety of receptors are characteristic features of eosinophils.

Historically, eosinophils have been considered to exert...
beneficial effects in the host defense against infections and tumors. However, eosinophils also release eosinophil-derived toxic mediators. Therefore, the infiltration of abundant eosinophils into inflamed tissue has been associated with detrimental effects in eosinophilic inflammation such as AR, eosinophilic chronic rhinosinusitis (ECRS) that was proposed as a new model for allergic rhinitis.

---

**Fig. 1. Features of Eosinophils: Receptors in Eosinophils, Eosinophil-Derived Mediators, and Multiple Biological Aspects of Eosinophils**

APRIL, a proliferation-inducing ligand; CCL, CC-chemokine ligand; CCR, CC-chemokine receptor; CR, complement receptor; CTH2, chemotractant receptor-homologous molecule on Th2 cells; CXCL, CXC-chemokine ligand; CXC, CXC-chemokine receptor; ECP, eosinophil cationic protein; ECRS, eosinophilic chronic rhinosinusitis; EDN, eosinophil-derived neurotoxin; EETs, eosinophil extracellular traps; EGF, epidermal growth factor; EGE, eosinophilic gastroenteritis; EGIDs, eosinophilic gastrointestinal diseases; EoE, eosinophilic esophagitis; EP, prostaglandin E; EPO, eosinophil peroxidase; FcR, Fc receptors; GM-CSF, granulocyte-macrophage colony-stimulating factor; ICAM-1, intercellular adhesion molecule-1; IDO, indoleamine, 2,3-dioxygenase; IFN, interferon; IL, interleukin; KYN, kynurenine; LFA-1, lymphocyte function-associated antigen-1; LT, leukotriene; MBP, major basic protein; NGF, nerve growth factor; NOD, nucleotide-binding oligomerization domain protein; PAF, platelet-activating factor; PD, D-prostanoid; PDGF, platelet-derived growth factor; PD1, protectin D1; PIRB, paired immunoglobulin-like receptor B; PPAR-γ, peroxisome proliferator-activated receptor-γ; PRR, pattern-recognition receptor; PSGL1, P-selectin glycoprotein ligand 1; RAGE, receptor for advanced glycation end-products; R, receptor; ROS, radical oxygen species; RIG-1, retinoic acid-inducible gene I; TGF, transforming growth factor; TSLP, thymic stromal lymphopoietin; Tx, thromboxane; SCF, stem cell factor; VIP, vasoactive intestinal peptide; VLA, very late antigen; TLR, Toll-like receptor; TNF, tumor necrosis factor; TXB, thromboxane; VIP, vasoactive intestinal peptide; VLA, very late antigen; 15-HETE, 15-hydroxyeicosatetraenoic acid. (Color figure can be accessed in the online version.)
clinically diagnosed phenotype of chronic rhinosinusitis with nasal polyp, eosinophilic otitis media, asthma, atopic dermatitis, eosinophilic gastrointestinal diseases (EGIDs), hyper eosinophilic syndromes (HES), and eosinophilic myopathies (13) (Fig. 1). Surprisingly, Leckie et al. reported that in a double blind, randomized, placebo-controlled trial, a monoclonal antibody against IL-5 (essential for the formation of eosinophils) did not affect the late asthmatic response or airway hyper responsiveness (AHR) to histamine. In addition, it prevented the increase in the number of eosinophils in both the bloodstream and sputum following allergen challenge, suggesting that eosinophils did not directly contribute to the development of remodeling, fibrosis, and AHR.8)

Subsequently, in 2004, two interesting studies of a murine ovalbumin-induced asthma model using different lines of eosinophil-deficient mice were reported by Lee et al.9) and Humbles et al.10) The former targeted the depletion of eosinophils using an eosinophil-specific promoter, the EPO gene, to drive the expression of a cytotoxic protein diphtheria toxin A (termed PHIL mice).11) In the latter, mice harbored a deletion of the high-affinity transcriptional factor GATA-binding site in the GATA-1 promoter (termed Δdbl-GATA mice).10) In both types of eosinophil-deficient mice, the number of infiltrating eosinophils in the tissue and blood was not altered in the asthma model. Moreover, PHIL and Δdbl-GATA mice were protected from the development of AHR and airway remodeling, respectively, in a model of experimental asthma.9,10)

Thus, the involvement of eosinophils in the pathogenesis of asthma has been controversial due to the discrepancies and contradictory observations in animal and human diseases. This suggests that eosinophils may be spatiotemporally altered to have multiple biological functions depending on the environment.

2. BENEFICIAL ASPECTS OF EOSINOPHILS IN HOST DEFENSE

2.1. Antiinfection Effects

2.1.1. Parasitic Infections

The eosinophil is one of the innate immune cells and a critical player in host defense. In parasitic helminthic infections, eosinophils have been histologically recognized to possess distinctive antiparasitic activities and to exert those effects via eosinophil-derived granule proteins MBP and ECP.11,12) Sasaki et al. reported that depletion of eosinophils by treatment with an anti-IL-5 antibody prolonged the survival of Angiostrongylus cantonensis in mice.13) In contrast, in studies of a parasitic infection model using eosinophil-depleted mice in vivo, the manipulation of eosinophils had no significant impact on disease development during Schistosoma mansoni infection.14,15) Although the role of eosinophils in in vivo helminthic infection remains unclear, eosinophilia is a sign of helminthic infection and eosinophils are involved in host-protective effects during parasitic infections.

2.1.2. Bacterial Infections

The granule proteins MBP and ECP contribute to the bactericidal ability of eosinophils.16) Torrent et al. demonstrated that ECP binding to bacteria-wall lipopolysaccharides (LPS) and peptidoglycans induced an antibacterial effect following bacterial aggregation.17) Moreover, a recent study has found that eosinophil activation by LPS from Gram-negative bacteria releases mitochondrial DNA-containing granule proteins, similar in structure to neutrophil extracellular traps, which are networks of extracellular fibers (primarily composed of DNA from neutrophils) and can bind pathogens. These proteins also exhibit antimicrobial activity independent of eosinophilic death.18)

2.1.3. Fungal Infections

Yoon et al. reported that eosinophils inhibited Alternaria alternata by releasing cytotoxic granule proteins (e.g., EDN) and MBP through signaling interaction between the adhesion β2-integrin molecule (CD11b) expressed on eosinophils and β-glucan, which is a major cell wall component of fungi, in a contact-dependent manner.19) Regarding the mechanism involved in this process, Matsuwaki et al. showed that Alternaria aspartate protease activated human eosinophils through the protease-activated receptor-2, and its signaling induced the production of IL-8, release of granule proteins (MBP and EDN), and upregulation of CD11b and CD63 on the surface of eosinophils.20,21)

2.1.4. Viral Infections

Recently, it has been shown that eosinophils and eosinophil-derived mediators may play a role in promoting virus clearance and antiviral host defense against the parainfluenza virus and respiratory syncytial virus (RSV).22,23) In the initial studies, Domachowske et al. reported that eosinophils exerted an antiviral effect against RSV through the release of recombinant EDN from eosinophils.24) Moreover, Adamko et al. revealed that eosinophils induced through ovalbumin sensitization and challenge decreased viral loads during parainfluenza virus infection.25) Eosinophils recognize single-stranded RNA through the TLR-7–MyD88 pathway and provide MyD88-dependent host defense against RSV.26) The investigators reported that virus clearance from lung tissue was more rapid in hypereosinophilic (IL-5 transgenic) mice compared with wild-type mice.26)

2.2. Antiinflammatory Effects

Eosinophils exert an antiinflammatory effect through their enzymes, such as histaminase, acid phosphatase, collagenase, arylsulphatase B, phospholipase D, lysophospholipase, catalase, and nonspecific esterases.12) Recently, protectin D1 (PD1), a lipid mediator biosynthesized from the omega-3 fatty acid docosahexaenoic acid, has been identified as a potent agonist for the resolution of inflamed tissues.27) Yamada et al. found that activated eosinophils in the inflamed site produced PD1, promoting the resolution of acute peritonitis induced by zymosan A, and exerted antiinflammatory activity.28) In humans, Miyata et al. showed that eosinophils were a major source of PD1 in patients with asthma, and the decreasing synthesis of PD1 in eosinophils was impaired in patients with severe asthma.29) Collectively, these results suggest that the antiinflammatory effects of eosinophils are spatiotemporally modulated depending on the progression and degree of the disease.

2.3. Antitumor Effects

Eosinophilia is frequently observed in several types of cancer. In various types of solid tumors (e.g., colorectal, breast, oral, esophageal, nasopharyngeal, laryngeal, penile, bladder, and prostate cancer), tumor-associated tissue eosinophilia has been linked to improved prognosis. However, in sarcoma (Hodgkin’s lymphoma), it is associated with poor prognosis.28,31) In animal experiments using an eosinophil-deficient model, Simson et al. showed that tumor incidence was greatly increased in the total absence of
eosinophils. This finding supported the notion that eosinophils play a potential role as effector cells in tumor immune surveillance. In contrast, da Silva et al. reported that inflated eosinophil levels in the tissue were a risk factor for oral squamous cell carcinoma, suggesting that eosinophils may be responsible for the deleterious outcome of experimental tongue carcinogenesis.

Although the influence of eosinophilia on the prognosis of cancer remains controversial, it seems likely that eosinophils exert their antitumor effects by secreting a variety of cytokines and cytotoxic granule proteins, including ECP, EDN, and EPO. In this antitumor process, eosinophils initially migrate to the tumor through tumor- and T cell-secreted cytokines and chemokines, including IL-2, IL-3, IL-5, IL-25, eotaxin-1, thymus and activation-regulated chemokine, and high-mobility group box-1. Subsequently, these infiltrated eosinophils induce an antitumor effect through ECP, EDN, MBP, TNF-α, and granzyme. This occurs after the binding of eosinophils with the tumor using CD11a/CD18, 2B4, and natural-killer group 2, member D expressed on eosinophils and intercellular adhesion molecule (ICAM)-1, CD48, and major histocompatibility complex (MHC) class I chain related-proteins A/B expressed on tumor cells, respectively.

3. ASPECTS OF EOSINOPHILS DETRIMENTAL TO HOST DEFENSE

3.1. Differentiation of Eosinophils and Their Accumulation via Adoptive Th2 and Innate Th2 Responses in Eosinophilic Inflammation

3.1.1. Differentiation of Eosinophils

Regarding the development of eosinophils, recent studies have clarified that transcription factors (e.g., GATA-1, PU.1, and CCAAT/enhancing binding protein) are involved in the process of eosinophil differentiation and maturation. In Th2-pathogenesis (e.g., AR and asthma), eosinophils are commonly differentiated from eosinophil progenitor cells (EoPs) expressing CD34 and IL-5Rα in bone marrow (BM) by growth factors and proinflammatory cytokines (e.g., IL-3, IL-5, and granulocyte macrophage colony-stimulating factor) mediated by Th2 lymphocytes and eventually migrate into the bloodstream. Of note, eosinophil lineage-committed progenitor cells have also been found in the cord blood, peripheral blood, BM, lung tissue, and sputum following allergic response. However, it remains unknown whether these in situ progenitors are actually generated, differentiate into eosinophils, and proliferate in tissue. Subsequently, these differentiated eosinophils from EoPs are generally recruited into the tissue through interactions between selectins and integrins on endothelial cells and adhesion molecules on eosinophils via chemokine gradients from Th2 cells. Finally, the activated tissue eosinophils exert their effects.

3.1.2. Accumulation of Eosinophils via Adoptive Th2 and Innate Th2 Responses

Asthma and ECRS have several phenotypes and are heterogeneous disorders. Therefore, it has been challenging to explain the accumulation of eosinophils in the tissues of patients with ECRS and/or asthma using only the classical Th2 pathway through adaptive immunity following an allergen-specific IgE-mediated response. Interestingly, recent studies have classified innate lymphoid cells (ILCs) into three major types: type 1 (natural killer cells, ILC1, cytotoxic T (Tc)1, and Th1); type 2 (ILC2, Tc2, and Th2); and type 3 (ILC3, Tc17, and Th17). In particular, ILC2 regulates type 2 immune responses by producing large amounts of Th2 cytokines prior to the acquired immune response. Moreover, several reports revealed that eosinophils are not only recruited by Th2 lymphocyte-derived IL-5 but also through ILC2-derived IL-5 stimulation with IL-1β, IL-18, IL-25, IL-33, and thymic stromal lymphopoietin from epithelial cells, fibroblasts, and mast cells.
macrophages following stimuli (e.g., parasites, fungi, protease allergens, and viruses). Figure 2 shows the pathways via adoptive Th2 and the innate Th2 response upon accumulation of eosinophils.

3.2. Activation Markers and Subtypes of Eosinophils in Inflammation

3.2.1. Markers of Eosinophil Activation

Although eosinophils can be counted in the blood and are a minor (<5%) component of circulating leukocytes, larger numbers of eosinophils occur in tissue outside the vasculature. Activation of eosinophils in the tissue is commonly implicated in the development of eosinophil-associated diseases. In an analysis of murine eosinophil maturation and activation in different stages (e.g., fetal liver, BM, blood, spleen, and infected lung), Voehringer et al. reported that the expression levels of C–C motif chemokine receptor 3, sialic acid-binding immunoglobulin-like lectin-F (Siglec-F), FIRE, CD62L, and phorhadius insect-related-A/B were altered during the distinct stages. Markers of eosinophil activation (e.g., CD69, L-selectin, ICAM-1 (CD54), CD44, P-selectin glycoprotein ligand-1 (CD162), cytokine receptors, Fc receptors (FcRRII), and integrins (CD11b)) have been proposed (Fig. 1). However, markers of eosinophil activation in inflamed sites in humans have not been sufficiently analyzed thus far, owing to the difficulty in obtaining adequate numbers of human eosinophils from biological samples.

Recently, we have reported that increased expression of CD69 on activated tissue eosinophils in ECRS was significantly correlated with nasal obstruction and the degree of pulmonary dysfunction. CD69 signaling induced the release of EPO from eosinophils, suggesting that CD69 on eosinophils is not merely a biomarker linked to clinical findings but also a potential therapeutic target for patients with ECRS and asthma (in press).

3.2.2. Subtypes of Tissue Eosinophils

Eosinophils have been classified according to morphological change, including differences in density and/or surface expression markers. Based on morphological changes, these cells have been classically divided into hypodense and normodense eosinophils, reflecting released and contained rich granules, respectively. Hypodense eosinophils have been recognized as activated eosinophils following the release of mediators and dominantly observed in allergic inflammation. Based on surface markers, Mesnil et al. have recently reported two distinct types, resident eosinophils expressing Siglec-FhighCD62LlowIL-3Rhigh and recruited inflammatory eosinophils expressing Siglec-FlowCD62LhighCD101highCD26LlowIL-3Rlow in the steady-state and mite-induced airway allergy lung, respectively. Moreover, they also reported that human resident eosinophils (Siglec-8CD62LIL-3Rlow) and inflammatory eosinophils (Siglec-8CD62LIL-3Rhigh) were found in nonasthmatic lungs and in the sputa of eosinophilic asthmatic patients, respectively.

Based on previous reports, Abdala-Valencia et al. suggested a tissue-based classification of murine eosinophils, specifically addressing four different types: EoP eosinophils (immature eosinophils recruited as precursors); steady-state eosinophils (tissue residents in morphogenetically quiescent tissues); type 1 eosinophils (innate defense and transient morphogenetic contexts); and type 2 eosinophils (Th 2 immune response, typically found in epithelial contexts). Collectively, this evidence suggests that there may be several subtypes of eosinophils, as shown in Fig. 2.

3.3. Eosinophilic Airway Inflammation

In upper and lower airway inflammation including AR, ECRS, and asthma, infiltration of eosinophils is a common feature of the inflammatory response; eosinophils infiltrate the airway tissue from the bloodstream mainly through cytokines and chemokines. The degree of eosinophilia is genetically correlated with the disease severity and exacerbation frequency of asthma.

Although numerous groups have reported that the accumulation of activated eosinophils promotes the pathogenesis of eosinophilic airway inflammation through eosinophil-derived toxic mediators (i.e., granules, cytokines, and chemokines), the exact involvement of eosinophils in this process remains controversial because of the discrepancies and contradictory observations between the mouse model and human disease. Nevertheless, activated tissue eosinophils contribute to the development of: 1) remodeling and fibrosis; 2) AHR; 3) mucus hypersecretion; and 4) dysregulation of airway nerves, reflecting the cardinal features of eosinophilic airway inflammation.

3.3.1. Remodeling and Fibrosis

Eosinophils release multiple growth factors and fibrogenic mediators that induce airway remodeling, such as transforming growth factor-β (TGF-β), matrix metalloproteinase-9, tissue inhibitor of metalloproteinase-1, vascular endothelial growth factor, basic fibroblast growth factor, angiogenin, specific granule proteins (MBP, EDN, and ECP), IL-13, IL-17, heparin-binding-epidermal growth factor-like growth factor, nerve growth factor, cytokine receptors, and integrins (CD11b). Ohno et al. reported that TGF-β, implicated in tissue remodeling via fibroblast proliferation and increased production of collagen and glycosaminoglycans, is mainly produced by eosinophils in bronchial biopsies obtained from asthmatic patients.

3.3.2. AHR

Using eosinophil-deficient mice in an asthma model, it was reported that eosinophils contribute to the formation of AHR. Gundel et al. found that the administration of MBP directly into the trachea of primates resulted in a significant increase in AHR with methacholine inhalation in a dose-dependent manner, while MBP and EPO induced transient bronchoconstriction immediately after instillation. Although ECP and EDN did not induce these effects, serum EDN and ECP (other markers of eosinophilic inflammation) may aid in the evaluation of the severity and bronchial hyperresponsiveness of childhood asthma.

3.3.3. Mucus Hypersecretion

Eosinophils also induce mucus hypersecretion from mucus cells through eosinophil-derived mediators (e.g., leukotrienes, IL-13, and ECP), supporting the finding that eosinophil deficiency prevented hyperproduction of mucus production in the asthma model. Recently, several interesting reports have demonstrated that activated human eosinophils in tissue release extracellular chromatin to form DNA traps through cytolytic extracellular trap cell death. Ueki et al. reported that the local cytolysis of eosinophils releases ECP and generates nuclear-derived DNA traps that were major extracellular structural components of eosinophil-rich secretions. Similar to neutrophil extracellular traps, these eosinophil extracellular traps contribute substantially to mucus plugging and the
vicosity of eosinophil-rich exudates that are involved in the exacerbation of ECRS or allergic bronchopulmonary aspergillosis. 3,62

3.3.4. Dysregulation of Airway Nerves

Upper airway inflammation, such as AR and/or ECRS, is frequently accompanied by lower airway inflammation (e.g., asthma). In the clinical setting, upper and lower airway inflammation is often termed “united airway disease” or “one airway, one disease,” and eosinophils seem to play an important role in this concept. 63 Interestingly, treatment of ECRS also improves asthma, suggesting three possible mechanisms: 1) a decrease in postnasal drainage of inflammatory mediators from the upper to the lower airways; 2) a reduction in systemic mediators disseminated by the upper airway; and 3) neural modulation via the nasal–bronchial reflex (NBR). 63 Although the behavior of eosinophils in the NBR remains unclear, eosinophils induce bronchoconstriction through effects on both sensory afferent and parasympathetic efferent pathways, reflecting control airway reflexes. 64

In addition, the loss of muscarinic (M2) receptor function by eosinophil–released MBP potentiates parasympathetic nerve-mediated bronchoconstriction, since MBP is an allostERIC antagonist of parasympathetic nerve M2 receptors. 65 Moreover, a reduction in the number of airway eosinophils using an anti-IL5 antibody prevented M2 receptor and airway function in the asthma model. 66 Previous reports demonstrated that physical interactions between lymphocyte function-associated antigen-1 on eosinophils and ICAM-1 on airway parasympathetic nerves led to the development of airway hyperreactivity and excessive bronchoconstriction. 67,68 Those findings suggested that the contribution of eosinophils to neuroinflammation, accompanied by airway nerve remodeling, is a key mechanism of increase in AHR in eosinophilic airway inflammation. 64,69

3.4. Other Eosinophil-Associated Diseases As shown in Fig. 1, eosinophils are also associated with the pathogenies of HES, EGID, inflammatory bowel disease (Crohn’s disease and ulcerative colitis), lung disease (idiopathic pulmonary fibrosis, pulmonary hypertension, acute lung injury, and chronic obstructive pulmonary disease), atopic dermatitis, and demyelination diseases (multiple sclerosis and neuromyelitis optica).

3.4.1. HES

The involvement of eosinophils in HES was first reported by Chusid et al. in 1975. 70 It is characterized by hypereosinophilia defined as: 1) >1500 eosinophils/µL for >6 months; 2) absence of any known etiology (e.g., parasitic infection and allergic disorders); and 3) signs and symptoms of organ involvement in heart failure, gastrointestinal dysfunction, central nervous system abnormalities, fever, or weight loss. 70 Of note, attention is required for HES patients with cardiovascular complications due to high morbidity and mortality associated with these disorders. 71,72 In 2006, HES were classified by a working group into myeloproliferative, lymphocytic, or other forms, followed by revisions in 2010. 73,74 The myeloproliferative form is further divided into myeloproliferative HES and chronic eosinophilic leukemia; the former is defined as myeloproliferative disease without proof of clonality, while the latter is defined as clonal eosinophilia including Fip1-like-1/platelet-derived growth factor receptor-α fusion protein resulting in the clonal proliferation of eosinophils (positive chronic eosinophilic leukemia). 74 In contrast to the myeloproliferative form, the lymphocytic form develops from aberrantly activated T-cell clones that constitutively produce eosinophilopoietic cytokines (e.g., IL-5) and is further divided into the clonal T cell and the non-T-cell clone type. 74

3.4.2. EGIDs Including Eosinophilic Esophagitis and Gastroenteritis

In the digestive tract, eosinophils are normally found in the gastrointestinal tract and cecum but not in the esophagus, as well as in the lamina propria but not in Peyer’s patches and intraepithelial locations in the tissue. 1,5 Eosinophilic esophagitis (EoE) was first described by Landres et al. in 1978. 75 Currently, EGIDs are commonly divided into EoE and eosinophilic gastroenteritis, and their prevalence is increasing in Asia and Western countries. 76–78 EoE and eosinophilic gastroenteritis mainly cause dysphagia and food impaction with vague symptoms and presence of abdominal symptoms including pain and diarrhea, respectively. The pathogenesis of EGIDs is commonly induced by an allergic response to food or environmental allergens through eosinophil-derived mediators (e.g., eotaxin-3, IL-5, IL-13, TGF-β, and periostin), and reﬂuxed gastric acid may also play an important role in the exacerbation of EoE. 77,78

4. IMMUNOREGULATION

4.1. Immunoregulation by Eosinophils

Eosinophils also modulate the immune response through interaction with proinflammatory cells (e.g., T cells and mast cells), eosinophil-derived mediators (e.g., granule proteins), various immunoregulatory cytokines, and lipid mediators. 1,5 For example, eosinophil-derived MBP, ECP, EPO, and nerve growth factor activate mast cells, resulting in the release of histamine. 79,80 EDN promotes the maturation and activation of dendritic cells. 81–83 MBP upregulates IL-8 production, superoxide release, and complement receptor 3 expression of neutrophils. 84 Antigen-specific interaction between MHC class II-expressing eosinophils and T cells results in an increase in T-cell proliferation and the production of cytokines from T cells. 85,86 Chemokine (C–C motif) ligands 17 and 22 induce the recruitment of Th2 cells. 87 Furthermore, the proliferation-inducing ligand (APRIL) and IL-6 are implicated in plasma cell survival, while IL-4 and IL-13 maintain alternatively activated macrophages (AAMs) in adipose tissue. 89

4.2. Th1/Th2 Modulation by Eosinophils

Eosinophils secrete cytokines (IL-2, IL-4, IL-6, IL-12, IL-10) that can promote T-cell proliferation and activation of Th1 or Th2 polarization. 90 Woerly et al. reported that eosinophils from hyper eosinophilic patients expressed CD86 and CD28, and CD28 signaling induced IL-2 and IFN-γ secretion by eosinophils, whereas there was no secretion of IL-4, IL-5, or IL-10. 91 In addition, in a mouse eosinophil-induced airway inflammation model, we reported that intratracheally instilled eosinophils following Th2 activation also resulted in an increase in Th1 cytokine (e.g., IFN-γ) expression derived from eosinophils, accompanied by AHR and lung inflammation. 92 That finding suggested that eosinophil-derived IFN-γ is an important mediator of lung inflammation which also modulates the Th1/Th2 balance. 92,93 Recently, in an extensive genome-wide association study of asthmatic patients in Western countries, it has been reported that lung dysfunction correlates with the
activity of Th1 rather than Th2 pathway genes. Moreover, Steinke et al. showed that patients with aspirin-exacerbated respiratory disease, along with exuberant infiltration of eosinophils, were distinguished by the prominent expression of IFN-γ, and a substantial amount of IFN-γ as well as other cytokines was derived from eosinophils. Thus, this evidence suggests that eosinophil-derived cytokines may modulate the Th1/Th2 balance and are involved in the development of heterogeneous diseases with mixed Th1 and Th2 activation profiles. However, additional investigations are required.

5. HOMEOSTATIC FUNCTIONS AND EOSINOPHIL HOMEOSTASIS

5.1. Homeostatic Functions

5.1.1. Metabolic Homeostasis

Both metabolic syndrome and obesity are risk factors of cardiometabolic diseases, and obese patients are at increased risk of developing asthma. Epidemiological studies investigating the relationship between obesity and asthma reported that obese individuals with a body mass index of 30 kg m² per m² have a 92% increased risk of asthma. Moreover, Desai et al. showed that, unlike sputum eosinophils, bronchial submucosal eosinophils were elevated in obese patients with severe asthma, suggesting that eosinophilic inflammation may play an important role in this subset of patients. In adipose tissue, AAMs induced by IL-4 and IL-13, also termed M2 macrophages, maintain glucose homeostasis and are related to eosinophils. Wu et al. showed that resident eosinophils mediate macrophage differentiation into the AAM phenotype by eosinophil-derived IL-4 in white adipose tissue of mice. Furthermore, they demonstrated that mice fed a high-fat diet exhibited impaired glucose tolerance and insulin resistance in the absence of eosinophils, while helminth-induced adipose eosinophilia enhanced glucose tolerance. Those results suggested that resident eosinophils play an unexpected role in metabolic homeostasis through the maintenance of adipose AAMs associated with glucose homeostasis and offer protection against the development of type 2 diabetes. In addition, Molofsky et al. reported that ILC2s sustained visceral adipose tissue eosinophils and AAMs, and this effect was enhanced during intestinal helminth infection. Collectively, those results indicate that eosinophil-regulated AAMs in adipose tissue may play an important role in metabolic homeostasis.

5.1.2. Maintenance of Plasma Cells

Chu et al. demonstrated that eosinophils in BM were a major source of plasma cell survival factors, such as APRIL and IL-6. Notably, the number of plasma cells in the BM of eosinophil-deficient mice was much lower in both the steady state and state of immunization with T-cell-dependent antigen 2-phenyl-oxazolone (phOx), suggesting that eosinophils are essential for the long-term maintenance of plasma cells in the BM. Subsequently, they reported that activation of eosinophils enhanced the expression of plasma cell survival factors (APRIL, IL-6, IL-4, IL-10, and TNF-α) both in vitro and in vivo. Furthermore, they showed that eosinophils maintained IgA-expressing plasma cells in the intestinal lamina propria, suggesting that eosinophils are important determinants of immune homeostasis in gut-associated tissues.

5.1.3. Thymic Development

The presence of eosinophils in the thymus is implicated in the selection of T cells. Throsby et al. demonstrated that CD11c⁺ eosinophils were preferentially recruited during class I-restricted T-cell selection, suggesting an immunomodulatory role for eosinophils upon reaching the steady state. Thus, the eosinophilic component of the thymic microenvironment may participate directly in the selection of T cells or aid in the scavenging of dead cells that fail negative selection. Kim et al. showed that selective depletion of eosinophils led to impaired clearance of apoptotic cells in the thymus.

5.1.4. Mammary Gland Development and the Female Genital System

Eosinophils are also associated with the development of the mammary gland and female genital system. Eotaxin-regulated eosinophils with macrophages are required in the postnatal mammary gland. In addition, eosinophils are involved in the postpartum repair of tissue and tissue remodeling in human endometriosis.

5.2. Eosinophil Homeostasis

The involvement of eosinophils in homeostasis in the human blood was first mentioned by Domarus in 1931. In 1953, Halberg et al. reported that part of the periodic intrinsic mechanisms for the circadian variation in blood eosinophils in mice might be regulated by endocrine factors from the adrenal gland. In 1975, Pauly et al. revealed that meal timing, rather than the light–dark cycle, is involved in the eosinophil rhythm in mice. Currently, there is limited knowledge regarding the mechanisms of eosinophil homeostasis since its first description. However, Nussbaum et al. found that eosinophils were related to basal circadian oscillations through ILC2-derived IL-5, which is induced by the signaling of the circadian synchronizer vasoactive intestinal peptide through the vasoactive intestinal peptide receptor 2 on ILC2 after feeding. This evidence linked the number of eosinophils with metabolic cycling. Nussbaum et al. suggested that these biologic pathways are independent of intestinal microbiota and raised the possibility that helminthic parasites may have co-opted these fundamental pathways of host metabolic homeostasis.

6. CONCLUSION

Infiltrated eosinophils play a critical role in host defense through its innate immune response. However, they also exert a detrimental effect in the pathogenesis of eosinophilic inflammation through nonallergic as well allergic responses. Moreover, the immunoregulation induced by eosinophils further underlines that caution should be exercised when

activity of Th1 rather than Th2 pathway genes. Moreover, Steinke et al. showed that patients with aspirin-exacerbated respiratory disease, along with exuberant infiltration of eosinophils, were distinguished by the prominent expression of IFN-γ, and a substantial amount of IFN-γ as well as other cytokines was derived from eosinophils. Thus, this evidence suggests that eosinophil-derived cytokines may modulate the Th1/Th2 balance and are involved in the development of heterogeneous diseases with mixed Th1 and Th2 activation profiles. However, additional investigations are required.

5. HOMEOSTATIC FUNCTIONS AND EOSINOPHIL HOMEOSTASIS

5.1. Homeostatic Functions

5.1.1. Metabolic Homeostasis

Both metabolic syndrome and obesity are risk factors of cardiometabolic diseases, and obese patients are at increased risk of developing asthma. Epidemiological studies investigating the relationship between obesity and asthma reported that obese individuals with a body mass index ≥30 kg per m² have a 92% increased risk of asthma. Moreover, Desai et al. showed that, unlike sputum eosinophils, bronchial submucosal eosinophils were elevated in obese patients with severe asthma, suggesting that eosinophilic inflammation may play an important role in this subset of patients. In adipose tissue, AAMs induced by IL-4 and IL-13, also termed M2 macrophages, maintain glucose homeostasis and are related to eosinophils. Wu et al. showed that resident eosinophils mediate macrophage differentiation into the AAM phenotype by eosinophil-derived IL-4 in white adipose tissue of mice. Furthermore, they demonstrated that mice fed a high-fat diet exhibited impaired glucose tolerance and insulin resistance in the absence of eosinophils, while helminth-induced adipose eosinophilia enhanced glucose tolerance. Those results suggested that resident eosinophils play an unexpected role in metabolic homeostasis through the maintenance of adipose AAMs associated with glucose homeostasis and offer protection against the development of type 2 diabetes. In addition, Molofsky et al. reported that ILC2s sustained visceral adipose tissue eosinophils and AAMs, and this effect was enhanced during intestinal helminth infection. Collectively, those results indicate that eosinophil-regulated AAMs in adipose tissue may play an important role in metabolic homeostasis.

5.1.2. Maintenance of Plasma Cells

Chu et al. demonstrated that eosinophils in BM were a major source of plasma cell survival factors, such as APRIL and IL-6. Notably, the number of plasma cells in the BM of eosinophil-deficient mice was much lower in both the steady state and state of immunization with T-cell-dependent antigen 2-phenyl-oxazolone (phOx), suggesting that eosinophils are essential for the long-term maintenance of plasma cells in the BM. Subsequently, they reported that activation of eosinophils enhanced the expression of plasma cell survival factors (APRIL, IL-6, IL-4, IL-10, and TNF-α) both in vitro and in vivo. Furthermore, they showed that eosinophils maintained IgA-expressing plasma cells in the intestinal lamina propria, suggesting that eosinophils are important determinants of immune homeostasis in gut-associated tissues.

5.1.3. Thymic Development

The presence of eosinophils in the thymus is implicated in the selection of T cells. Throsby et al. demonstrated that CD11c⁺ eosinophils were preferentially recruited during class I-restricted T-cell selection, suggesting an immunomodulatory role for eosinophils upon reaching the steady state. Thus, the eosinophilic component of the thymic microenvironment may participate directly in the selection of T cells or aid in the scavenging of dead cells that fail negative selection. Kim et al. showed that selective depletion of eosinophils led to impaired clearance of apoptotic cells in the thymus.

5.1.4. Mammary Gland Development and the Female Genital System

Eosinophils are also associated with the development of the mammary gland and female genital system. Eotaxin-regulated eosinophils with macrophages are required in the postnatal mammary gland. In addition, eosinophils are involved in the postpartum repair of tissue and tissue remodeling in human endometriosis.

5.2. Eosinophil Homeostasis

The involvement of eosinophils in homeostasis in the human blood was first mentioned by Domarus in 1931. In 1953, Halberg et al. reported that part of the periodic intrinsic mechanisms for the circadian variation in blood eosinophils in mice might be regulated by endocrine factors from the adrenal gland. In 1975, Pauly et al. revealed that meal timing, rather than the light–dark cycle, is involved in the eosinophil rhythm in mice. Currently, there is limited knowledge regarding the mechanisms of eosinophil homeostasis since its first description. However, Nussbaum et al. found that eosinophils were related to basal circadian oscillations through ILC2-derived IL-5, which is induced by the signaling of the circadian synchronizer vasoactive intestinal peptide through the vasoactive intestinal peptide receptor 2 on ILC2 after feeding. This evidence linked the number of eosinophils with metabolic cycling. Nussbaum et al. suggested that these biologic pathways are independent of intestinal microbiota and raised the possibility that helminthic parasites may have co-opted these fundamental pathways of host metabolic homeostasis.

6. CONCLUSION

Infiltrated eosinophils play a critical role in host defense through its innate immune response. However, they also exert a detrimental effect in the pathogenesis of eosinophilic inflammation through nonallergic as well allergic responses. Moreover, the immunoregulation induced by eosinophils further underlines that caution should be exercised when
considering alterations in the Th1/Th2 balance by antibodies (e.g., IFN-γ) as therapeutic tools. Previous evidence suggested that these eosinophil-associated diseases must be managed by understanding the multiple aspects of eosinophils in response to environmental conditions. In addition, understanding the role of eosinophils in homeostasis may reveal interesting new therapeutic strategies against metabolic syndrome.

Acknowledgments This work was supported by funding from the Academic Society for Research in Otolaryngology, Kansai Medical University; a research grant from the Kansai Medical University Research Consortium; the Ministry of Education, Culture, Sports, Science and Technology (MEXT)-Supported Program for the Strategic Research Foundation at Private Universities (S1201038); Grants-in-Aid for Scientific Research (C) from MEXT (15K10793 and 19K09860); and the Branding Program as a World-Leading Research University on Intractable Immune and Allergic Diseases.

Author Contributions A.K., Y.Y., D.V.B., K.S., S.S., Y.K., M.A., and H.I. were involved in drafting the manuscript. All authors provided final approval for the publication of this manuscript and read and approved the final version.

Conflict of Interest The authors declare no conflict of interest.

REFERENCES


29) Miyata J, Fukunaga K, Iwamoto R, Isobe Y, Nium K, Takamiya...


