



Neutrophil activation in occupational asthma

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Purpose of review

The aim of this review is to emphasize the role of neutrophils in patients with occupational asthma. This review facilitates a better understanding, accurate diagnosis, and proper management of asthmatic reactions provoked at the workplace.

Recent findings

Increased recruitment and infiltration of neutrophils are found in patients with occupational asthma. Activated neutrophils release several mediators including pro-inflammatory cytokines and extracellular traps, leading to stimulation of airway epithelium and other inflammatory cells.

Summary

New insights into neutrophils in the pathogenesis of occupational asthma may provide a novel approach to the individual patient with occupational asthma.

Keywords

airway epithelium, non-Th2 inflammation, oxidative stress

INTRODUCTION

At the workplace, a latency period of exposure to causative agents (sensitization) and an acute re-exposure to the agents (inhalation) induce immunologic reactions and airway obstruction called occupational asthma [1]. There have been more than 300 agents inducing occupational asthma. However, isocyanates, drug powder, wheat flour, and grain dust are prevalent causes of occupational asthma [2^{*}]. These agents are commonly classified into two groups: high molecular weight (HMW) agents which lead to immunologic reactions including IgE-mediated response as a complete antigen and low molecular weight (LMW) agents which bind to tissue proteins to work as a complete antigen and induce immunologic/nonimmunologic mechanisms [3]. In addition, eosinophilic inflammation is induced by type 2 inflammation in response to causative agents; however, neutrophilic inflammation is also noted in patients with occupational asthma because of some agents [4–6]. There have been several studies to suggest the involvement of activated neutrophils in the airway inflammation [7^{*}]. In this review, we summarized recent data demonstrating neutrophil activation in the pathogenic mechanism of airway inflammation in occupational asthma, especially isocyanate (a representing phenotype of LMW agents)-induced occupational asthma and grain dust/wheat flour (representing phenotypes of HMW agents)-induced

occupational asthma, to extend our understanding and treatment strategies.

PATHOGENIC MECHANISMS OF NEUTROPHILS IN OCCUPATIONAL ASTHMA

Toluene diisocyanate-induced occupational asthma

Isocyanates are highly volatile and reactive chemicals widely encountered in various workplaces [8], among which toluene diisocyanate (TDI) is most frequently found in the manufacture of polyurethane foams, sealants, elastomers, adhesives, and coatings, and is the most prevalent cause of occupational asthma in Korea and worldwide [9,10]. TDI binds to tissue proteins to form conjugates, leading to immune responses in asthmatic airway [11], although various immune mechanisms of TDI-induced occupational asthma (TDI-OA) have not been completely understood.

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KEY POINTS

- In occupational asthma, environmental exposure to various agents at the workplace induces chronic airway inflammation and remodeling via various immunologic and nonimmunologic mechanisms.
- Although eosinophilic inflammation is a prominent feature in the airway of occupational asthma, neutrophilic airway inflammation is found in patients with occupational asthma caused by some agents such as isocyanate and grain dust.
- An increase in neutrophil infiltration and activation is associated with stimulation of airway epithelium, leading to the production of reactive oxygen species and inflammatory cytokines recruiting inflammatory cells including eosinophils and neutrophils.
- A novel molecule, extracellular traps, released from activated neutrophils could enhance activation of inflammatory cells including eosinophils and epithelial cells, contributing to airway inflammation refractory to anti-inflammatory therapy in patients with occupational asthma.

The finding of Th2-immune responses with eosinophilia was noted in the airway mucosa of TDI-OA patients [12]. Furthermore, emerging evidence has revealed that neutrophils are also involved in this asthmatic reaction. An immunohistochemical study demonstrated that neutrophil elastase-positive neutrophil counts were significantly higher in the airway mucosa of TDI-OA patients compared to that of allergic asthma [13]; levels of myeloperoxidase (MPO) and IL-8 in the sputum samples of TDI-OA patients significantly increased after TDI bronchial challenges, indicating that TDI-OA is associated with a prominent feature of neutrophilic airway inflammation [14,15]. TDI exposure further induces overproduction of matrix metalloproteinase 9, which enhances structural changes and neutrophil migration [16]. In addition, neutrophils decrease the production of clusterin/progranulin from airway epithelial cells, which enhances oxidative stress-induced epithelial damage and inflammation in patients with TDI-OA [17]. Furthermore, as an autoimmune mechanism, TDI binds to transglutaminase present on the surface of airway epithelial cells and induces epithelial cell activation, leading to persistent airway inflammation [18,19]. Considering autoimmunity, we further showed the effect of neutrophil extracellular traps (NETs; mesh-work of chromatin with bounded granule proteins) released by activated neutrophils on the expression of transglutaminase, cytokeratin, and α -enolase located in airway epithelial cells, which induce immune responses in the airway mucosa of TDI-OA [20,21].

It has been demonstrated that NETs prime immune cells and damage epithelial cells, resulting in an induction of airway inflammation [22]. Taken together, these findings suggest that neutrophils may contribute to the progression of airway inflammation in patients with TDI-OA.

Grain dust/wheat flour-induced occupational asthma

Workers in the food and farm industries are mainly exposed to flour and grain dust containing lipopolysaccharides [23,24]. Th2/IgE-mediated mechanisms have been believed to be critical in the pathogenesis of baker's asthma [25]. IL-4 is the key cytokine to promote IgE production/Th2 cell differentiation in allergic inflammation [26]; we reported a significant association between work-related respiratory symptoms (WRs) and the polymorphisms of IL-4 receptor α at Ile75Val A > G [27]. However, elevated levels of MPO, IL-8, and TNF- α were also found in the sera of bakery workers [28]. Moreover, neutrophil infiltration was enhanced in the airway mucosa of patients with grain dust asthma [29] and serum neutrophil chemotactic activity was increased after bronchial challenge tests with grain dust [30], suggesting that both Th2 and non-Th2 immune mechanisms are important in these types of occupational asthma. In addition, we investigated genetic associations of TLR4 with WRs in bakery workers and found that there were significant associations of WRs with the promoter polymorphisms at TLR4 at -2027 A > G and -1608 T > C [31]. TLR4 expressed in immune cells including eosinophils and neutrophils has been shown to play a crucial role against wheat flour and grain dust considering that lipopolysaccharide is contained in dust [32]. A recent study demonstrated that patients with baker asthma had a significantly higher level of serum mannose-binding lectin, according to genetic polymorphisms of the MBL2 gene [33]. Mannose-binding lectin is a member of the collectin family that plays an important role in innate defense by complement activation [34]. This pattern-recognition molecule is regulated by reactive oxygen species (ROS) produced by neutrophils and airway epithelial cells [35], indicating that innate immune responses with neutrophil activation contribute to developing WRs in grain dust/wheat flour-exposed workers.

DIAGNOSIS, MANAGEMENT, AND PERSPECTIVES

Diagnosis

A diagnosis of work-related asthma (occupational asthma or work-exacerbated asthma) among adult

asthmatics is based on comprehensive history taking, physical examination, and objective confirmation of the presence of asthma. Patient history includes the onset or worsening of asthma symptoms at work with exposure to common allergens and/or irritants, which improve at off-work with avoidance. It is also needed to obtain information on material exposure. Sputum eosinophilia/neutrophilia and changes in neutrophils with work-shifts or bronchial challenge tests may be helpful in accurate diagnosis. In cases combined with Th2-mediated occupational asthma, allergy skin testing and detection of serum specific IgE antibodies are supportive to confirm the causative agent; however,

these two methods are practical only for some agents [36]. Specific inhalation challenge testing with causative agents is the reference standard that is recommended for confirmative diagnosis. Airway hyperresponsiveness has been evaluated; however, some patients do not have airway hyperresponsiveness and may return to a normal condition after leaving the workplace [37]. Although increased neutrophil counts or levels of MPO/IL-8 in sputum samples may support a diagnosis of occupational asthma with neutrophil activation, they are not validated yet [38–40]. Further in-vitro and in-vivo methods will be needed to diagnose this type of occupational asthma.

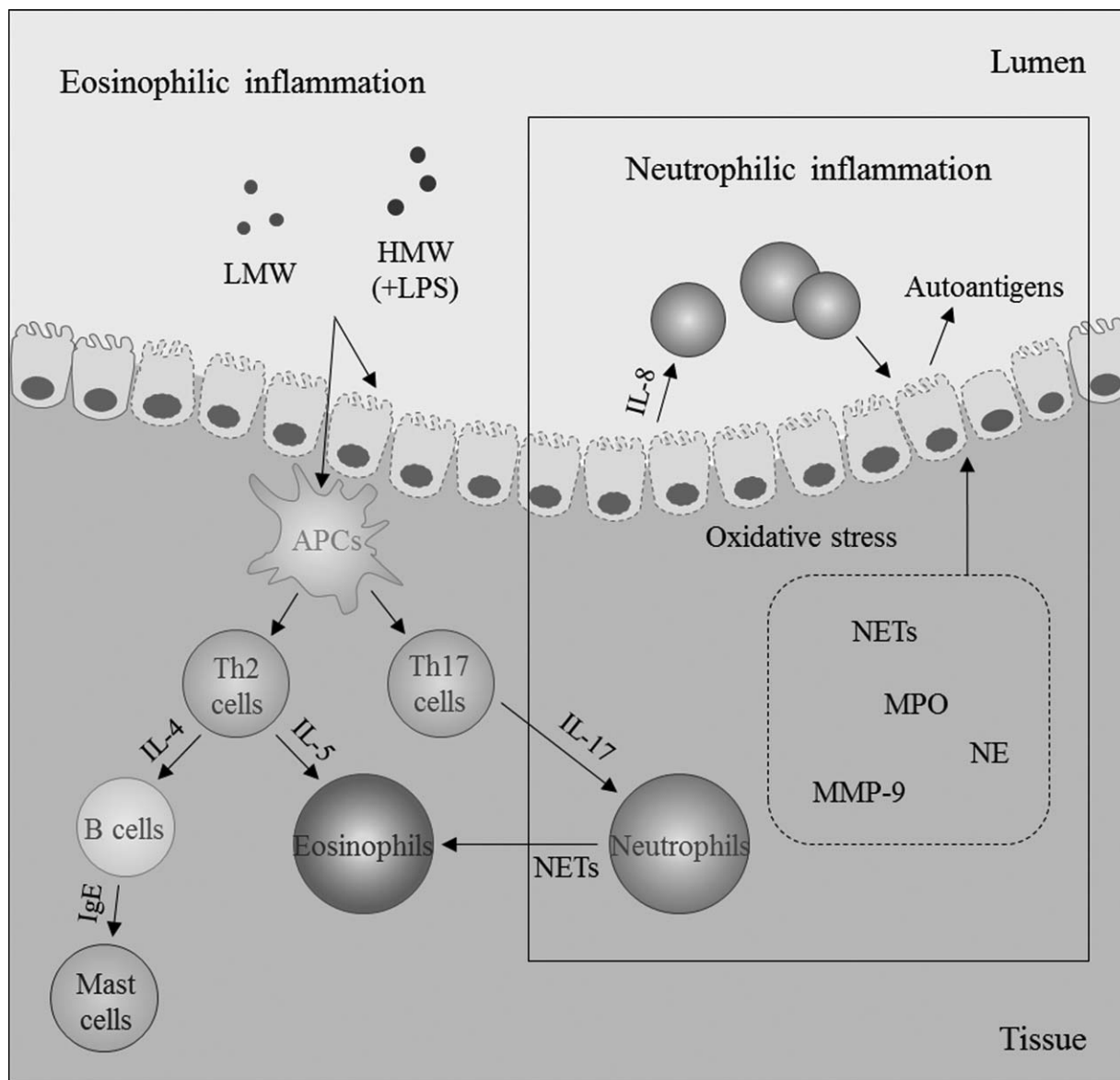


FIGURE 1. Schematic mechanisms of eosinophils and neutrophils possibly involved in occupational asthma. APCs, antigen-presenting cells; HMW, high molecular weight; LMW, low molecular weight; LPS, lipopolysaccharide; MMP-9, matrix metalloproteinase 9; MPO, myeloperoxidase; NE, neutrophil elastase; NETs, neutrophil extracellular traps.

Management and perspectives

Complete avoidance of exposure to causative agents is a basic step in the management of occupational asthma. Pharmacologic treatment of occupational asthma is similar to that of nonoccupational asthma. A recent study using cluster analysis documented that increased sputum neutrophil counts were associated with more severe asthma phenotypes [41]. Neutrophilic airway inflammation in asthmatic patients does not respond to inhaled corticosteroids, representing anti-inflammatory drugs for asthmatic patients [42]. As there have been emerging evidence of neutrophils being involved in the airway inflammation of occupational asthma as well as nonoccupational asthma, new treatment modalities that aim neutrophilic inflammation will be helpful in the better management of occupational asthma [43]. Th17 cells that secrete IL-17 are most extensively studied, contributing to neutrophilic inflammation. There was a strong correlation between IL-17 levels and neutrophil counts in the sputum and blood of neutrophilic asthmatics [44]. An in-vivo study revealed that lymphocytes exposed to dexamethasone had no inhibitory effect on IL-17 production [45]. Recently, brodalumab, a humanized monoclonal antibody to the IL-17 receptor, was investigated in moderate-to-severe asthmatics. Although it did not affect the primary outcome (the Asthma Control Questionnaire scores) of the study, it showed a positive effect in the subgroup of the study patients with highly reversible lung functions [46]. It was proposed that neutrophil autophagy and neutrophil extracellular traps could damage airway epithelium and facilitate inflammation of airway epithelial cells and peripheral blood eosinophils, which increases asthma severity [47^{***}]. As ROS plays an important role in neutrophil activation of occupational asthma including baker's asthma and TDI-OA, antioxidants have been suggested as a novel therapeutic approach in in-vitro models [48]. However, there has been no evidence whether antioxidants have beneficial effects in this type of occupational asthma.

CONCLUSION

The pathogenesis of occupational asthma caused by multiple agents remains largely uncertain. However, a recent study showed that neutrophilia is frequently found in workers exposed to occupational allergens. Activated neutrophils release several mediators such as pro-inflammatory cytokines and extracellular traps, leading to immune cell stimulation and airway epithelial damage involved in airway inflammation (Fig. 1). In addition, neutrophilic asthma is more severe and less responsive to

steroids compared with the eosinophilic phenotype. Therefore, further studies are needed to understand the role of neutrophils in patients with occupational asthma for the better management of the condition.

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Conflicts of interest

There are no conflicts of interest.

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