

Dangerous allergens: innate immunity, dendritic cells and allergic asthma

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Immune responses can be compartmentalized into innate versus adaptive components. This relatively recent dichotomy positioned the innate immune system at the interface between the host and the external environment, and provided a new conceptual framework with which to view allergic diseases, including asthma. Among the cells of the innate immune system, antigen-presenting dendritic cells are now thought to be intimately involved in allergen recognition, as well as modulating allergic immune responses. This review summarizes current thinking regarding the role of dendritic cells in allergic asthma and concludes with a summary of emerging concepts in the field.

KEYWORDS: adaptive immunity • allergy • asthma • danger signal • dendritic cell • innate immunity

The compartmentalization of immune responses into innate versus adaptive components was an important recent advance in cellular immunology [1]. As opposed to adaptive immunity, which is the major provenance of B and T lymphocytes, innate immune responses can theoretically encompass many other cell types that can respond to infections or tissue injury. Dendritic cells (DCs) are a crucial component of the innate immune system because they sense and respond to diverse environmental cues. Key distinguishing features between these two arms of the immune system include the patterns or antigens recognized, the kinetics of activation and the capacity for memory. In contrast to adaptive immune cells, which recognize an extremely broad repertoire of antigens due to genetic recombination of antigen-receptor genes, innate immune cells respond to a more limited set of targets via a conserved set of pattern recognition receptors (PRRs). Similarly, innate immune cells generally respond quickly to activating signals and have limited capacity for long-term memory. Signals that activate the innate immune system include those encoded by infectious pathogens or produced as a result of tissue injury or inflammation.

There has been an explosion of research in this area in recent years, with new PRR–ligand interactions being discovered at a rapid pace [2–7]. PRRs tend to be ancient and conserved, and probably evolved to recognize invading

pathogens. For example, the Toll-like receptor (TLR) family recognizes a diverse family of pathogen-encoded patterns, and evolved at least 100 million years before the rearranged antigen receptors that underlie adaptive immunity. In an exciting new area of research, TLRs have now been firmly implicated in allergic diseases [8] and are being exploited as novel adjuvants in allergen immunotherapy and other vaccine strategies. Some TLR–ligand pairs potentially have protective effects in allergen immunotherapy (e.g., TLR9 and CpG [9]). Other TLR ligands have been linked with allergen sensitization, although the dose and timing of exposure are critical modulating factors (e.g., TLR4 and lipopolysaccharide [LPS]).

In addition to recognizing cell wall components, nucleic acids and carbohydrates derived from microbes, innate immune receptors respond to an array of endogenous danger signals that indicate tissue injury. Examples of these endogenous ligands (or ‘alarmins’ [7]) include heat-shock proteins [10], extracellular matrix components (e.g., low-molecular-weight hyaluronic acid [11]), extracellular ATP [12], high-mobility group box-1 proteins [13], modified lipoproteins (e.g., oxidized low-density lipoprotein [14]), complement factors [15] and uric acid crystals [16]. This rapidly expanding world of endogenous danger signals has both conceptual and therapeutic implications. First,

since activation of innate immune receptors generally results in tissue inflammation (due to signals that attract and activate other immune cells), and inflammation-associated tissue injury will generate endogenous PRR ligands, there must be robust negative regulation of PRR-initiated signaling to prevent a vicious cycle from spiraling out of control. Although still a relatively new field of study, there is a growing body of literature identifying negative regulators of innate immune responses, some of which may be new therapeutic targets [17–21]. An interesting twist in this area came from the recent study by Kim *et al.* [22]. These investigators found that, in the absence of T and B cells, viral infection or challenge of mice with poly I:C (a TLR3 ligand) resulted in much more severe inflammation and higher mortality, both of which were inhibited by adoptive transfer of CD4⁺ or CD8⁺ lymphocytes [22]. This interesting study implies that adaptive immune cells can nonspecifically inhibit innate immune-driven inflammatory responses in mice.

A second implication of the expanding world of endogenous danger signals relates to the possibility of identifying shared signaling intermediates as novel therapeutic targets. For example, the Nod-like receptor-based inflammasome links innate immune exposure to various bacteria or toxins with caspase-1-dependent processing of cytokines, including IL-1 α , IL-18 and IL-33 [23,24]. Interestingly, the inflammasome was recently demonstrated to be required for activity of the proallergic adjuvant alum [25], but its involvement in sensitization to *bona fide* allergens remains to be determined. Since both IL-18 and IL-33 can contribute to allergic inflammation [26,27], targeting components of the inflammasome (e.g., NALP3) may hold therapeutic promise in allergic diseases. Future studies may also uncover signaling modules involved in different forms of inflammation. For example, could there be an innate immune signature characteristic of allergic inflammation that results in tissue eosinophilia, mast cell activation and tissue remodeling? It will be interesting in future studies to dissect specific innate immune signals derived from the innate immune system in asthma and other allergic diseases, which may help define qualitatively the different forms of inflammation [28].

Dendritic cells as environmental sensors

Several innate immune cell types can initiate or modulate immune responses to inhaled allergens, including DCs, airway epithelial cells and alveolar macrophages. DCs in particular are uniquely poised to respond to tissue injury or infection and alert adaptive immune cells to allergens and other danger signals [29]. Three features of lung DCs make them uniquely poised to be key environmental sensors. First is their anatomic location within the respiratory epithelium. Certain DC subsets may constitutively sample the airway lumen as part of an immunosurveillance system [30], whereas other DC subsets reside just below the surface epithelia and come in contact with external pathogens or antigens that either directly or indirectly disrupt the epithelial barrier. The second key feature of DCs is their ability to respond to diverse environmental cues. This is due to their expression of an array of cell-surface and intracellular PRRs, as well to their enormous capacity for endocytosis. The third key feature of DCs is their

ability to intimately interact with antigen-specific T cells. This is due not only to the migratory properties of these two cell types, which bring them into close contact in secondary lymphoid organs and in tissues, but also to the formation of an intimate DC–T-cell synapse that allows a two-way exchange of information. Thus, DCs provide a key link between exposure to inhaled allergens and the development of adaptive immune responses.

In the case of naive T cells, DC–T-cell interactions are thought to occur primarily in draining lymph nodes after activated DCs have migrated into the afferent lymphatics. In this location, allergen-bearing DCs will come into contact with naive T cells that recirculate through lymph nodes after emigrating from the vasculature via the high endothelial venules. Precisely when and where naive allergen-specific T cells are educated in human asthmatics is not known, but it seems reasonable to postulate that this occurs in peribronchial lymph nodes early in life. DCs that emigrate from tissues can also re-enter the bloodstream and gain access to other organs, although the contribution of these DCs to immune responses remains controversial. It should be remembered that secondary lymphoid organs (e.g., lymph nodes and the spleen) contain populations of nonmigratory follicular DCs that continually sample the afferent lymph. These resident DCs will take up cell-free soluble antigens that enter lymphatic channels, which will occur during vaccination or other conditions associated with a breach of epithelial barriers. Could inhaled allergens travel as immune complexes in afferent lymph and be retained in follicular DCs, which might sustain IgE production or other humoral responses? Future studies investigating the precise mechanisms by which allergens gain access to secondary lymphoid organs, and initiate and sustain allergen-specific immune responses in asthma, should be worthwhile.

Dendritic cell ontogeny

Dendritic cells derive from hematopoietic precursors and are a remarkably malleable cell type. A comprehensive discussion of DC ontogeny is beyond the scope of this review, and there have been excellent reviews of this topic recently [31–33]. It is becoming appreciated that both myeloid and lymphoid progenitors can give rise to DCs, with differing phenotypes depending on the precise nature of the differentiation signals encountered. Conventional DCs (cDCs) can be distinguished from plasmacytoid DCs (pDCs) owing to functional differences in these subsets and also to their differential expression of PRRs [31]. The ability to precisely distinguish between inflammatory monocytes and cDCs is becoming increasingly difficult, and it is probably best to view these two cell types at each end of a spectrum starting from CD14⁺ circulating monocytes and ending with canonical tissue myeloid DCs. Studies in mice point to a suppressive effect of pDCs in models of allergic airway inflammation [34], but it is currently unclear if human lung pDCs have a similarly suppressive function in asthma. Part of the difficulty in this regard relates to obtaining large numbers of human lung DCs for analysis. Furthermore, there is no single cell-surface marker that reliably identifies pDCs, which requires complex multicolor flow-cytometry gating strategies. Interestingly, human pDCs (such

as basophils) express cell-surface IgE, and recent studies have uncovered crosstalk between FcεRI and TLR-signaling pathways in these cells [35,36].

It is generally accepted that cDCs, especially those that differentiate from myeloid precursors, play an important and pro-inflammatory role in allergic asthma. Four lines of evidence support this contention. First, in mouse models of allergic airway inflammation, depletion of CD11c⁺ cDCs attenuated both the initiation of allergic sensitization as well as ongoing airway inflammation [37]. Second, adoptive-transfer studies in mice found that cDCs could recapitulate many features of allergic airway inflammation [37]. Taken together, these two observations indicate that CD11c⁺ cDCs are necessary and sufficient for allergen-driven airway inflammation, at least in mice. Third, studies of asthmatic human subjects have generally found that the numbers and activation state of respiratory tract DCs are increased compared with control subjects, and although these generally have a conventional phenotype, it is difficult to compare between different studies due to differences in study design (e.g., before and after allergen challenge vs unchallenged subjects) and immunolabeling protocols [38–40]. Fourth, therapeutic agents used in asthma, including inhaled glucocorticoids, leukotriene antagonists, anti-IgE agents and even β-agonists, can suppress DC activation both *in vivo* and *in vitro* [41].

Dendritic cell maturation

A current paradigm in immunology is that DCs reside in the peripheral tissues in an immature state, where they transduce an inhibitory signal that suppresses activation of T cells (including autoreactive clones that escaped thymic deletion) [42]. After activation by appropriate danger signals, DCs undergo a complex maturation process involving simultaneous changes in endocytosis, antigen processing and presentation, and secretion of pro-inflammatory and immunoregulatory cytokines and chemokines. DC maturation also results in changes in chemokine receptor expression that facilitate homing to regional lymph nodes where they present peptide antigen in the context of MHC (signal 1), as well as costimulatory molecules (signal 2) to naive T cells. The idea that 'educated' DCs (similar to T cells) can also recirculate in the bloodstream and migrate to other organs (e.g., bone marrow and the thymus [43]) is gaining attention, although the frequency of these cells is very low and their contribution to immune responses remains controversial.

Epithelial and other stromal cells are thought to influence DC maturation and their subsequent ability to activate T cells [44]. DCs integrate these stromal signals in a way that influences their ability to instruct both T-cell homing and T-cell lineage commitment (i.e., signal 3). This is a new and exciting area of immunology, where very little is known in human allergic diseases. Experiments in mice clearly show that signals imprinted in DCs during their maturation induce the expression of tissue-specific homing receptors in responding T cells [45]. In the case of skin Langerhans' cells, this involves upregulation of chemokine receptors and adhesion molecules on T cells (e.g., C–C chemokine receptor [CCR]-10 and cutaneous lymphocyte antigen [CLA]) that bind to their respective ligands

expressed in inflamed skin (e.g., C–C chemokine ligand [CCL]-27 and E- and P-selectin) [46]. In the case of gut-educated DCs, this involves upregulation of chemokine receptors and adhesion molecules on T cells (e.g., CCR9 and the integrin α4β7) that bind to their respective ligands expressed in inflamed skin (e.g., CCL25 and MadCAM-1) [47]. Although it seems probable that similar receptor–ligand pairs will govern T-cell homing to the lung, to date, there is little firm evidence in support of this notion. One interesting possibility is that allergen sensitization through the skin would result in atopic dermatitis (AD) because CLA⁺ allergen-specific memory would be established, whereas sensitization through the respiratory tract results in asthma (or rhinitis) by a similar mechanism. The observation that many subjects with AD have a dysfunctional skin epithelium due to genetic and acquired defects in filaggrin and other barrier proteins provides strong support for this possibility [48,49]. Whether this 'barrier defect' also exists in asthma is an interesting and active area of research that provides a new framework to investigate allergen sensitization and crosstalk between the innate and adaptive immune systems [50]. Although this model is attractive, it is difficult to study prospectively in human subjects and would require a cohort study investigating blood and tissue DCs and T cells over time.

Dendritic cells shape T-helper immune responses

In addition to affecting T-cell-homing patterns, DCs also shape CD4⁺ Th-cell differentiation. By contrast to Th1-promoting DCs, which secrete high amounts of IL-12 family members, the molecular mechanisms by which DCs induce differentiation of other T-cell lineages (e.g., Th2 and Th17) are not as well understood. DCs do not produce IL-4, the critical Th2-promoting cytokine. One early hypothesis was that Th2-promoting DCs arose by 'default' in the absence of strong TLR signals. This was thought to reflect 'low-grade' DC activation, sufficient to upregulate costimulatory molecules but not enough to induce robust secretion of Th1-promoting cytokines, such as IL-12 or IL-23. This provided a cellular and molecular correlate of the hygiene hypothesis, although it is now clear this default model is an oversimplification and that DCs can actively influence Th2-lineage differentiation [41]. DCs and other cells can promote Th2 responses but contact-dependent or soluble signals are still to be identified, although some candidates have recently emerged. These include secreted products (e.g., thymic stromal lymphopoietin [TSLP] [51–53]) and DC surface markers (e.g., OX40L and members of the Jagged family of cell-surface receptors [54–56]) [57]. Since the original description by Soumelis *et al.*, TSLP has now been identified as an important epithelial-derived pro-Th2 factor that may also be involved in Th2 memory [58]. TSLP is produced by keratinocytes [59] but in numerous studies of human and mouse DCs exposed to house dust mites, ragweed allergen extract and other danger signals *in vitro*, we have not detected secretion of TSLP using sensitive ELISA techniques, despite the induction of 'pro-Th2' DCs (data not shown). Thus TSLP is not always necessary for the generation of a pro-Th2 DC phenotype, but it may be important in tissues when allergens and other inhaled particles contact epithelial cells.

We postulated the existence of an environmental 'signal X' that induced the maturation of immature DCs into a Th2-promoting phenotype, and speculated that this signal would either be encoded in, or interact with, allergens. We recently showed that ambient particulate matter (APM; i.e., particulate air pollution) fulfilled the function of 'signal X' because APM-exposed DCs promoted a Th2-like pattern of cytokine secretion in responding T cells (see later). Because APM-exposed DCs were robustly activated and secreted IL-12 and other cytokines, this argued against a 'default' model of DC activation. Thus, exactly how pro-Th2 DCs instruct Th2-cell differentiation is currently unclear, and there may be more than one mechanism in this regard.

Allergens can activate tissue & circulating innate immune cells

Allergens themselves can directly activate different cell types, including airway epithelial cells. The house dust mite *Dermatophagoides pteronyssinus* (Der p) are probably the best studied in this regard, and these mites are now known to contain discrete allergens, many of which have enzymatic activity. These include the class 1 cysteine protease Der p 1 and the class 3 serine protease Der p 3 [60]. A seminal early paper reported that Der p disrupted tight junctions of the airway epithelium in a protease-dependent manner [61], suggesting a mechanism by which inhaled allergen would gain access to innate immune cells in the airway. In addition to house dust mites, other allergens or allergen extracts have been shown to directly activate epithelial cells and other structural cells (e.g., fibroblasts), including ragweed, cockroach and mold allergen (Pen C 13) [62–66]. Pichavant and colleagues showed that Der p 1 stimulated epithelial monolayers to secrete chemokines, including CCL20, which induced DC chemotaxis using a transwell coculture system [67]. CCL20 (or MIP-3 α) is a potent chemoattractant for immature DCs because immature DCs (e.g., naive T cells) express the CCL20 receptor CCR6. Interestingly, we recently reported that CCL20 is elevated in bronchoalveolar lavage (BAL) fluids following segmental allergen challenge and correlated strongly with BAL lymphocytes, although we did not enumerate DCs in that report [68]. Thus, CCL20 may be a sentinel chemokine that alerts and recruits innate and adaptive immune cells to the lung, and is an attractive therapeutic target in allergic asthma. Hematopoietic cells can also be targeted by allergens. In an interesting recent paper, Sokol *et al.* demonstrated that basophils were essential in eliciting Th2-driven immune responses in mice using papain as a model allergen and protease [28]. By contrast, Page *et al.* recently found that cockroach allergen ('frass') caused allergic airway inflammation and activated neutrophils in mice in a TLR2-dependent manner [69]. Thus, allergens appear to induce proallergic immune responses via multiple innate immune pathways.

Cysteine proteases: signaling mechanisms & potential therapeutic targets?

The cysteine protease activity of Der p 1 appears to be essential for its proallergic properties. For example, Kikuchi *et al.* recently showed that mice immunized with 'protease-dead' Der p 1

(induced either with the irreversible inhibitor E-64 or by heat denaturation) did not mount IgE responses when immunized by intraperitoneal injection with alum [70]. Immunization with E-64 did not affect ovalbumin-driven IgE responses, suggesting that it was specifically targeting Der p [70]. One possibility is that the protease activity of Der p 1 was required for 'pro-Th2' DC activation, but this has not been formally investigated to date. Studies with Der p were reminiscent of the cysteine protease CPB2.8 from *Leishmania mexicana*, which was found to be a potent inducer of Th2 responses [71]. Interestingly, Corry *et al.* suggested that protease activity is the defining feature of 'class II' allergens, including extracts of *Aspergillus* (i.e., those that require neither priming nor adjuvants for their effects) [72]. Taken together, these studies strongly support the idea that allergens and (cysteine) protease activity are intimately linked.

Two general mechanisms have emerged for the protease-dependent effects of Der p and these are not mutually exclusive. First, Der p 1 can cleave several relevant cell-surface molecules, including CD23 [73], CD25 [74] and CD40 [75]. This mechanism may be especially apparent after preactivating Der p *in vitro* with cysteine. Interestingly, Shahib *et al.* showed that dust mite allergen Der p 1 inhibited IL-12 secretion in LPS-stimulated DCs by cleaving CD40 expression [75]. Second, Der p can activate the protease-activated receptor (PAR) family of G-protein coupled cell-surface receptors [76–78]. PAR1 was the first member of this family identified and was found to be the main thrombin receptor [79]. Different PAR family members have now been associated with allergic diseases and asthma, and PAR agonists/antagonists are being developed therapeutically [80]. Allergens appear to activate PAR family members in a cell-type specific manner, and there is currently a lack of consensus in this area. For example, Adam *et al.* reported that Der p 1 activated A549 cells in a PAR2-independent manner, and induced ERK1/2- and nuclear factor (NF)- κ B-dependent IL-8 expression [77]. By contrast, another study suggested that Der p 1 activated PAR2 but not PAR1 in the same cell line [81]. Cockroach allergen was found to stimulate PAR2 on fibroblasts [62], whereas the mold allergen Pen C 13 activated epithelial cells via both PAR1 and PAR2 [63]. PAR2 may be particularly important for Th2-immune responses and allergic diseases [82]. For example, PAR2 is upregulated in human asthmatic epithelium [83] and PAR2 knockout mice are partially protected from asthma [84], in association with reduced IL-4 secretion [85]. Interestingly, PAR2 appears to be required for DC development from bone marrow in mice [86]. Although it is expressed on keratinocytes and monocytes, a comprehensive understanding of the role of PAR2 and other related family members in allergic immune responses is lacking. Taken together, these studies suggest that strategies that counteract protease activity and signaling may have novel antiallergic effects, and that defective antiproteases may be novel susceptibility genes in allergic diseases.

Other allergens also directly activate DCs

In addition to Der p, other allergens have now been shown to directly influence DC differentiation, even in the absence of epithelial-derived signals (FIGURE 1). For example, Traidl-Hoffmann

et al. showed that phytoprostanes associated with birch pollen inhibited DC IL-12 production [87], whereas Shreffler *et al.* reported that Ara h 1 was a ligand for DC-SIGN [88]. We have been studying how short ragweed extract (RWE) affects DC differentiation and found that RWE potently activates mouse bone marrow-derived DCs in an oxidative stress-dependent manner (RANGASAMY T, UNPUBLISHED DATA). This is in keeping with the observation that ragweed and other pollens have intrinsic pro-oxidant activities [66], and adds to the idea that oxidative stress can be sensed as 'dangerous' by the innate immune system [14].

Proallergic adjuvants

In addition to signals intrinsic to allergens themselves, real-world exposure to airborne allergens will occur together with inhaled particles or other biologically active components that can activate DCs. These nonallergenic components could act as natural or man-made adjuvants that shape subsequent adaptive immune responses. Indoor allergens, including dust mite and cockroach frass, will probably contain endotoxin and other TLR ligands from bacteria colonizing the arthropod digestive tract [89]. Endotoxin is a potent activator of DCs that binds TLR4 and initiates Myd88-dependent signaling and DC maturation. It is also a powerful adjuvant whose ability to influence the Th1/Th2 balance appears to be critically dependent on its concentration; high concentrations induce robust IL-12-secreting DCs that cause Th1 differentiation, whereas low concentrations induce IL-12-secreting DCs that instead promote Th2 differentiation [90]. Although the idea that 'a little bit' of endotoxin is pro-Th2 is becoming widely accepted, two caveats are worth bearing in mind. First, the landmark study by Eisenbarth *et al.* that established this idea used different doses of inhaled LPS in a mouse model of allergic airway inflammation [90]. Inhaled LPS will induce local airway inflammation and may not be directly binding DC TLR4 in these models. Second, there is currently little biochemical evidence that LPS induces qualitatively different TLR4-dependent signals in any cell type.

In urban areas, outdoor aeroallergens complex with airborne particulate matter (PM), which is the respirable fraction of air pollution whose composition varies based on source and location. The idea that PM is a potential man-made 'proallergic adjuvant' has been around since pioneering studies by Diaz-Sanchez and others [91], but its effects on DCs have only recently begun to be investigated. PM is categorized according to size, and particles range from coarse (defined as ~10 µm in aerodynamic diameter) to ultrafine (defined as <100 nm). In most developed countries, the levels of outdoor PM are strictly regulated since PM exposure is now strongly linked with exacerbations of cardiac and

pulmonary diseases. In support of its potential role as an inhaled adjuvant, there are growing bodies of epidemiologic data linking PM exposure and asthma incidence. PM size has major effects on its aerodynamic dispersal as well as deposition in the respiratory tract, with smaller particles reaching deeper into the lung than larger coarse PM, which will be largely filtered by the upper airways. Thus, depending on the precise nature of the particles to which they are absorbed, aeroallergens will differentially impact on the upper or lower airways and, thus, reach different cells of the respiratory innate immune system.

Particle composition can also have important effects on target cells, including DCs. For example, Porter *et al.* showed that PM that was enriched either in car or truck exhaust had distinct effects on human monocyte-derived DCs *in vitro* [92]. Williams *et al.* found that DCs exposed to ambient outdoor PM (from Baltimore, MD, USA) induced a distinct profile of DC maturation, characterized by enhanced expression of costimulatory molecules and cytokines [93]. In coculture with allogeneic naive CD4⁺ T cells, Baltimore PM-exposed DCs augmented T-cell IL-13 secretion and promoted a mixed Th2/Th1 cytokine profile. This PM was enriched in heavy metals and transition elements that induced DC oxidative stress [93]. Future studies investigating the precise mechanisms by which inhaled particles contact lung DCs and initiate their maturation *in vivo* should prove worthwhile.

Expert commentary

The positioning of the innate immune system at the interface between the host and the external environment provides a new framework with which to view allergic diseases and anti-allergic

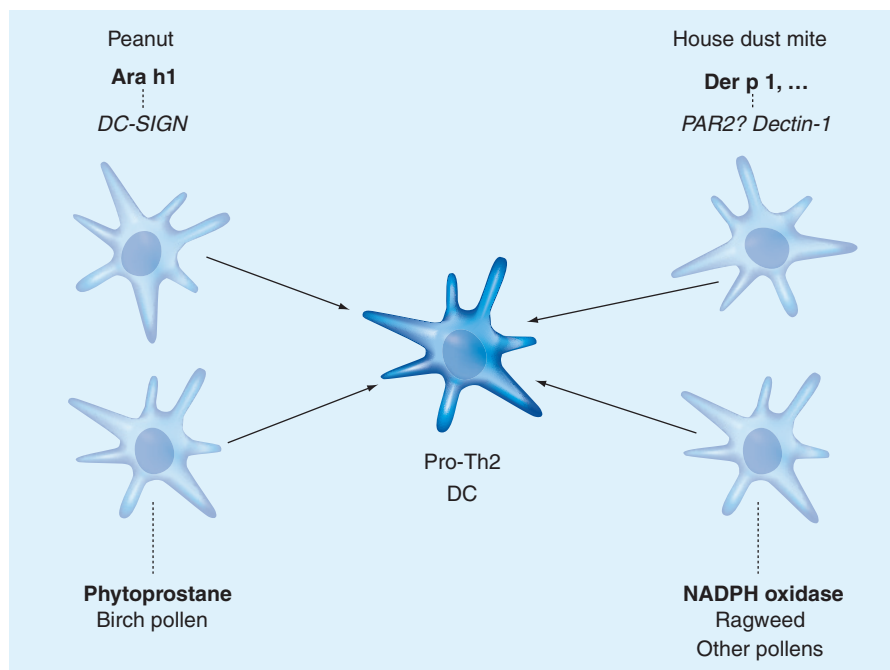


Figure 1. Potential mechanisms whereby allergen-encoded signals could directly activate immature dendritic cells and induce their maturation into a Th2-promoting phenotype (dark cell). Allergen-associated molecules are depicted in bold and potential dendritic cell receptors are depicted in italics.

therapies. Innate immune receptor families and signaling cascades initiate and perpetuate allergic inflammation, and understanding how existing therapies impact these pathways will provide new insights into anti-allergy therapies. By eavesdropping on the cross-talk between epithelial cells, DCs and lymphocytes, we can gain insight into the pathophysiology of allergic diseases and identify future strategies for intervention.

Five-year view

The field of 'allergic innate immunity' is rapidly growing and attracting the interest of many laboratories and research groups. It seems probable that new allergen-PRR interactions will be uncovered and it may be possible to define an innate immune signature specific for different allergic diseases. New therapeutic targets should be forthcoming as a result of research into the molecular mechanisms of innate immune-receptor signal transduction, and it will be particularly interesting to follow research into negative regulation of innate immune receptors. These studies may uncover endogenous inhibitory compounds or pathways

that can also be exploited therapeutically. DC-based therapeutics in allergic asthma, either based on novel vaccination strategies or drug-delivery approaches, are a real possibility in the next 5 years. Technical advances in multicolor flow cytometry should facilitate the identification of different DC lineages in the lung, and may define subsets particularly important in promising sustained airway inflammation in asthma. Although fundamental new discoveries will continue to be achieved using mouse models, it will be important to remain alert to species-specific differences in innate immune-cell lineage and function, and expand human translational research in this field.

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Key issues

- The compartmentalization of immune responses into innate versus adaptive components was an important advance and provided a new conceptual framework to view allergic diseases.
- Dendritic cells (DCs) are key sentinel cells in the lung owing to their ability to sense and respond to diverse environmental cues.
- Owing to their intimate interactions with T cells, DCs provide a crucial link between exposure to allergens and subsequent adaptive immune responses.
- Allergens can activate innate immune cells using different receptors and signaling pathways, and either directly or indirectly influence DC differentiation.
- Many allergens, including *Dermatophagoides pteronyssinus*, activate innate immune cells in a protease-dependent manner, which can involve protease-activated receptors or cleavage of cell-surface molecules.
- Real-world exposure to allergens will occur in the context of other biologically active components and particles, including Toll-like receptor ligands and ambient air pollution.
- Defective epithelial barrier function may predispose susceptible subjects to allergen sensitization through the skin or respiratory tract.
- Defining the innate immune signaling mechanisms by which allergen exposure predisposes to allergic inflammation will enhance our understanding of the pathophysiology of asthma and should suggest new therapeutic targets.

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