Factors in clinical expression of allergic airways disease

Lopuhaä, C.E.

Citation for published version (APA):
CHAPTER 1

General Introduction
Allergy

Allergic sensitisation is a common feature with variability in clinical manifestations. It is characterised by an immediate hypersensitivity reaction in which allergen-specific IgE antibody plays a key role. These allergen-specific IgE antibodies are produced by B-type lymphocytes and regulated by interleukines (IL-4, IL-13)\(^1,2\), produced by allergen-specific T\(_{helper 2}\) lymphocytes\(^3\). During an allergic reaction the cross-linking of surface bound high affinity IgE receptors (Fc\(_e\)RI) on mast cells results in the release of various mediators, with proinflammatory properties\(^4\). The variety of symptoms that can develop after allergen exposure involves several organs. In this thesis the symptoms and signs of the airways in inhalant allergy are of main interest.

Epidemiology of inhalant allergy

Inhalant allergy is considered to be a major cause of bronchial hyperresponsiveness and asthma. In epidemiological studies there is a close association between allergy and asthma. Sensitisation to common inhaled allergens as detected in skin tests and radio allergosorbent test (RAST) is found in 40 to 50% of the population in industrialised countries\(^5\). The prevalence of allergic asthma however, is only 5-10 %\(^6\). The majority of individuals sensitised to inhaled allergens have upper airway symptoms like allergic rhinitis or even have no symptoms at all.

The manifestations of inhalant allergy like allergic rhinitis and allergic asthma depend on genetic and multiple environmental factors. These include airway exposure to allergens, endotoxins\(^7\), tobacco smoke and air pollutants\(^8\) as well as sibship size, socio-economic status, early childhood infections and dietary habits. The last decades have shown an increase in the prevalence of sensitisation to inhaled allergens and a parallel increase in allergic rhinitis and allergic asthma\(^5\). Several suggestions have been made to explain the growth in incidence of sensitisation to inhaled allergens. Since the peak incidence of all inhalant allergies is in the first decade of life, exposure to environmental factors in early life may have important impact on the development of the allergic diseases. Even intrauterine and neonatal factors were shown to be linked with increased serum IgE concentrations as well as to the development of asthma\(^9\).
lower body weight at birth was reported to be associated with an increased risk for asthma in adult life, although the latter finding remains controversial\textsuperscript{10}.

Despite the above-mentioned studies, a conclusive explanation for the recent increase in the incidence of allergy and asthma is still lacking.

Asthma and rhinitis often coexist\textsuperscript{6} and there appear to be similarities in the genetic background\textsuperscript{11}. Nevertheless, little is known what causes the diversification of the allergic phenotype into nasal and/or bronchial involvement.

The allergic reaction

The early phase of the allergic reaction
The reaction induced by inhaled allergens can be divided in an early- and late allergic response. Clinically, the early allergic reaction is defined as a response of the target organ like skin, nose or bronchi within 30 minutes after exposure to inhaled allergens. This reaction involves IgE-mediated activation of mast cells or basophilic granulocytes, induced by cross-linking of surface bound high affinity IgE receptors (Fc\textsubscript{e}RI). The mast cell is recognized to be the essential cell in the early response of the effector phase\textsuperscript{4}. The cross linking of high affinity receptors triggered by complexes of receptor-bound allergen specific IgE and allergens results in the release of inflammatory mediators like histamine, platelet activating factor, several leukotrienes and prostaglandin D\textsubscript{2}\textsuperscript{4}. These mediators are amongst others responsible for the vasodilatation, oedema and smooth muscle cell contraction. This leads to itching, sneezing, watery discharge and congestion in the upper airways\textsuperscript{12}, whereas bronchoconstriction in the lower airways causes wheezing and dyspnoea\textsuperscript{13}. The early reaction in the skin is characterised by erythema and oedema caused by vasodilatation and increased vascular permeability\textsuperscript{14}.

Activation of mast cells has been shown to result in synthesis and secretion of several cytokines, most of which are likely to be involved in the subsequent induction of the late phase of the allergic reaction.
The late phase of the allergic reaction

The distinction of the early and late phase of the allergic reaction became evident in studies using experimental allergen challenge in skin, nose and bronchi. During more or less continued exposure to inhaled allergens under natural conditions early and late phase mechanisms are frequently superimposed. The late phase of the allergic reaction is characterised by an influx of inflammatory cells and their release of mediators. This reaction generally starts about 3 to 8 hours after allergen exposure. Symptoms of the late allergic reaction are an erythematous induration in the skin and airway obstruction in the upper as well as the lower airways. Nasal blockage is due to vasodilatation and oedema of the nasal mucosa, bronchial obstruction is primarily caused by bronchial smooth muscle contraction.

Numbers of eosinophilic and neutrophilic granulocytes, macrophages and lymphocytes are increased in broncho-alveolar lavage fluid and bronchial biopsies obtained during the late phase after bronchial allergen challenge. Similarly, an increase in eosinophilic granulocytes and their mediators during the late phase after nasal allergen challenge has been found in late phase nasal lavages. This local eosinophilia during late allergic reactions is due to the recruitment from the bone marrow, induction of cell adhesion molecules and prolonged cell survival. These events are orchestrated by the cytokines IL-4, IL-5 and GSM-CSF, which are probably produced by activated Th2-lymphocytes and mast cells.

Methods to study allergic sensitisation and allergen induced inflammation

Radio allergosorbent test (RAST)

In the RAST the level of allergen specific IgE in serum is detected. Specific IgE in serum binds to the allergens that have been coupled to a solid phase (agarose or cellulose). Subsequently, radio-labelled anti-human IgE is added and binding of specific IgE can be detected. As van Toorenbergen et al. have shown, the level of circulating IgE is in free equilibrium with the cell-bound IgE, which is responsible for the immediate allergic reaction.
Skin Test
The outcome of both skin prick test and intracutaneous test depends on the mast cell numbers, the sensitivity of the mast cells for IgE mediated activation and the sensitivity of the cells of the target tissues for mast cell products. In both tests a small amount of allergen is applied into the skin which can induce a positive early cutaneous reaction within 30 minutes characterised by a wheal (oedema) and flare (erythema) reaction. A late cutaneous reaction is characterised by a less clearly defined erythematous skin induration which develops 3 to 8 hours after the early cutaneous reaction. The determination of the allergen threshold concentration, i.e. the lowest concentration of allergen which elicits a positive skin reaction, shows the highest correlation with the level of allergen-specific IgE antibodies.

Bronchial hyperresponsiveness
Bronchial hyperresponsiveness refers to an exaggerated response of bronchial smooth muscle cells to bronchoconstrictor stimuli. These stimuli can be non-specific non-sensitising physical or chemical stimuli, pharmacological agents like histamine or specific sensitising agents like inhaled allergens in sensitised individuals.

Bronchial hyperresponsiveness is a hallmark of asthma and is associated with asthma severity and need for medicinal support. However, it may also be found in non-asthmatic diseases like allergic rhinitis. The degree of bronchial hyperresponsiveness is the main determinant of the bronchial response to allergen and is associated with the late allergic bronchial obstruction. Bronchial hyperresponsiveness in allergic sensitisation to inhaled allergens is related with the degree of (sub)mucosal inflammation of the airway wall. Moreover, bronchial exposure to allergen in sensitised individuals induces an increase in bronchial hyperresponsiveness, which may persist for several weeks. So far it remains unclear whether bronchial hyperresponsiveness is caused by an increased sensitivity of the structural elements of the bronchial wall including bronchial smooth muscle cells or is mediated by the presence of inflammatory cells in the airway wall. The measurement of the degree of bronchial hyperresponsiveness ("direct challenge") is expressed as the concentration of the inhaled provocative agent (histamine or metacholine), which induces a 20% decrease of the forced expiratory volume in 1 second $[\text{FEV}_1]$. 

14 chapter 1
**Bronchial allergen challenge**

The model of human whole lung challenge has been used for many studies and has proven its value for studying the pathogenesis of allergic asthma\(^{17}\). In bronchial allergen challenge, increasing doses of allergen are applied via a dose aerosol in the lower airways. An early allergic bronchoconstriction occurs approximately 10 minutes after allergen inhalation, and generally resolves spontaneously within 90 minutes. This initial reaction is caused by an IgE-mediated mast cell activation, resulting in mediator release and smooth muscle contraction. The allergen dose that results in a standardised early asthmatic reaction (PD\(_{20}\)allergen) is strongly determined to bronchial hyperresponsiveness as established by histamine or metacholine\(^{38,45}\). There is a considerable overlap between asthma and non-asthmatic rhinitis in allergen threshold as well as in bronchial hyperresponsiveness to pharmacological agents like histamine\(^{46}\).

A second phase of bronchoconstriction may develop 3 to 4 hours after allergen inhalation and may last up to 48 hours. This late asthmatic reaction is considered to be related to inflammatory changes in the bronchial wall including cell influx, mucosal oedema and plasma protein exudation. These inflammatory changes occur in parallel with an increase in bronchial hyperresponsiveness and closely resemble the findings in chronic asthma\(^{18,20,47-49}\).

Bronchoscopy with broncho-alveolar lavage or bronchial biopsies has proven to be useful to study bronchial inflammation. More recently, it was shown that the less invasive technique of sputum induction by inhalation of hypertonic saline is able to provide similar information\(^{50}\).

**Nasal allergen challenge**

Allergic rhinitis is generally present in patients with allergic asthma. Allergen induced mucosal inflammation in the nose resembles allergen induced bronchial mucosal inflammation. Despite this resemblance, nasal allergen challenge is not frequently used as a model for allergen induced lower airway inflammation as seen in allergic asthma. Although the allergen induced cellular inflammation is similar in the bronchi and the nose, there is a considerable difference in structure of both organs\(^{51}\). The main difference is the presence of smooth muscle cells in the bronchi, which can be activated by mediators produced by mast cells and non-resident inflammatory cells. Smooth muscle cells are completely lacking in the nasal cavity\(^{52,54}\).
Similar to bronchial allergen challenge outcomes of nasal challenge can be measured functionally by change in upper airway resistance (rhinomanometry)\textsuperscript{55,56}. Inflammatory changes can be determined by non-invasive methods (nasal washings)\textsuperscript{56,57} as well as invasive methods (nasal brushes and biopsies)\textsuperscript{16,58,59}. The early reaction is induced by mast cell activation and followed by vasodilatation and triggering of sensory nerve endings\textsuperscript{60,61}.

The late allergic inflammation in the nose is not always accompanied by symptoms as the nasal congestion is generally mild during this phase\textsuperscript{62}. However, rhinomanometry during late allergic reaction is able to demonstrate a recurrence of nasal obstruction. Moreover, nasal sampling has shown a second increase in histamine release as well as inflammatory cell influx\textsuperscript{21,22,63,64}.

\textit{Nitric Oxide}

Nitric oxide in exhaled air (eNO) is associated with bronchial inflammation in asthma. Levels of eNO are increased in patients with asthma, and these levels are associated with severity of the disease as well as with bronchial hyperresponsiveness\textsuperscript{65-67}. The inducible form of the protein NO synthase (iNOS) is upregulated by proinflammatory cytokines and subsequently increases the production of NO\textsuperscript{68}. Allergen induced increase in bronchial inflammation is shown to be accompanied by an increase in eNO\textsuperscript{69,70,72}. Moreover, it has been demonstrated that treatment of asthma with inhaled corticosteroids reduces the inflammation in the lower airways and decreases the level of eNO\textsuperscript{73}. Based on these observations, NO in expired air is considered to be a marker for bronchial inflammation in asthma.
Aim of the thesis

The cascade of events leading to allergic airways disease can schematically be divided in (1) initial exposure to inhaled environmental allergens, probably early in life, (2) the subsequent production of IgE antibodies to these allergens (sensitisation), (3) the continued exposure to allergen after sensitisation, (4) the inflammatory processes after interaction between allergen and IgE antibodies, including activation of mast cells and recruitment and activation of other inflammatory cells and finally (5) the response of the structural elements of the target organ like smooth muscle cells in the bronchi. Theoretically, presence or absence of symptoms of a target organ in sensitised individuals may be determined by differences at any of the four final stages of this cascade.

It can be envisioned that allergic asthmatic patients have higher levels of specific IgE, higher levels of exposure to inhaled allergens, increased allergic inflammatory responses or increased sensitivity of target tissues for inflammatory mediators than their non-asthmatic allergic counterparts. Literature provides little support for the first two options. Occupational asthma in which high exposure to inhaled allergen is a decisive factor may be an exception in this respect. Difference in sensitivity of target tissues is probably an important factor in the development of allergic asthma. Reactivity of bronchial smooth muscle cells or their activation pathways is probably influenced by genetic background. In addition, environmental factors like viral infections and bronchial vascular hydrostatic conditions can influence airway smooth muscle responsiveness. The finding that a continued local allergic inflammatory reaction increases the sensitivity of the target tissues complicates the precise localisation of the difference between the asthmatic and non-asthmatic allergic phenotype. Chronic airway inflammation causes structural changes in the airways generally referred to as “airway remodelling”.

The majority of the studies described in this thesis are focussed on possible differences in the allergen induced inflammatory processes (level 4) between asthmatic and non-asthmatic allergic individuals. Allergen induced inflammation was studied in house dust mite allergic individuals with or without asthma. In order to investigate whether differences are specific for the bronchi or detectable in other target organs, allergen induced bronchial inflammation was compared with allergen induced nasal inflammation.
In *chapter 2* we investigate the effect of prenatal exposure to famine on the development of allergic sensitisation, lung function and on obstructive airways diseases in adult life. For this purpose lung function, total IgE and specific IgE against several inhaled allergens was measured in a large cohort that had been exposed to the Dutch famine in 1944-1945 during fetal life.

In *chapter 3* we describe the associations between parasite infections, IgE antibodies and skin reactivity to inhaled allergens in a large cohort of children in Gabon. It was investigated whether helminthic infections and infections with malarial parasites might play a role in the difference in prevalence of inhalant allergy between western industrialised countries and less developed nations.

*Chapter 4* describes a study in which bronchial allergen challenges were performed with isolated major allergens of house dust mite (Der p1, Der p 2) in allergic asthma. In this model of allergen-induced IgE-mediated bronchial responses we investigate the relationship between immediate bronchial responses, the degree of non-specific bronchial hyperresponsiveness and the level of sensitisation to house dust mite allergen.

In *chapter 5* we investigate whether there are differences in late phase allergen-induced bronchial inflammation between allergic asthma and non-asthmatic rhinitis. We performed whole lung challenges with house dust mite and compared changes in inflammatory parameters in induced sputum and in serum.

By analogy of the study described in chapter 5, in *chapter 6* we compare changes in allergic inflammatory parameters during the late phase of the allergic reaction in the skin and the nose in sensitised subjects with or without asthma.

The inducible form of Nitric Oxide synthase (iNOS) is associated with bronchial inflammation. In *chapter 7* we study the changes of levels of eNO in house dust mite allergic patients after a standardised early asthmatic reaction.
References


34. Witteman AM, Sjamsoedin DH, Jansen HM, van der Zee JS. Differences in nonspecific bronchial responsiveness between patients with asthma and patients with rhinitis are not explained by type and degree of inhalant allergy. Int Arch Allergy Immunol 1997;112:65-72.


22 chapter 1


