Curr Allergy Asthma Rep (2014) 14:435
DOI 10.1007/s11882-014-0435-7

RHINITIS (JJ OPPENHEIMER AND J CORREN, SECTION EDITORS)

Cytokine Profiles in Allergic Rhinitis

Guy Scadding

Published online: 16 March 2014

Abstract Allergic rhinitis, particularly seasonal allergic rhinitis, is considered a classic Th2-mediated disease, with important contributions to pathology by interleukins 4, 5 and 13. As such, allergic rhinitis is an excellent model for studying allergic inflammation, with findings potentially relevant to the mechanism of lower airways inflammation seen in allergic asthma. However, recent evidence has revealed roles for additional non-Th2 cytokines in asthma, including IL-17 family cytokines and epithelial-derived cytokines. Additionally, putative roles for epithelial-derived cytokines and innate lymphoid cells have been described in chronic rhinosinusitis with nasal polyps. Here, evidence for the involvement of different cytokines and cytokine groups in allergic rhinitis is considered.

Keywords Allergic rhinitis · Cytokine · Cytokine profile · Interleukin · Th2 · IL-17 · TSLP · IL-33

Introduction

Cytokines constitute a diverse group of immunomodulatory, signalling molecules with a wide range of functions in health and disease. Whilst initially identified to be of lymphocyte origin, they are now known to be produced by many different cell types, including immune, structural and organ-specific tissues. They orchestrate host responses to infection and trauma, as well as the potentially damaging responses seen in allergic, inflammatory and autoimmune disease.

Cytokines play an essential role in mediating allergic inflammation. The importance of the 'type-2'/Th2 cytokines, particularly IL-4 and IL-13, in both the development of allergic sensitisation and pathology of allergic inflammation in asthma is well established [1]. However, inflammatory responses in asthma are more complex than simple overexpression of Th2 cytokines. Recent research has identified additional contributions from the IL-17 family of cytokines [2] and epithelial-derived cytokines such as TSLP [3] and IL-33 [4], amongst others.

Allergic rhinitis, particularly seasonal rhinitis, is an excellent model for studying allergic inflammation, where the triggering factor(s) can clearly be identified, and sufferers can be studied during periods of disease and remission. Moreover, the nasal mucosa is easily accessible for provocation with allergen and recording of responses, both clinical and immunological. This article will discuss the cytokine profiles identified in studies of allergic rhinitis in humans. The evidence for Th2-predominant responses will be discussed, as well as roles for other cytokine groups, including Th1-, Th17- and epithelial-derived cytokines.

Approaches to Investigating Nasal Cytokines

Nasal secretions are produced by seromucous glands and goblet cells, with possible additional input from leaked plasma contents during acute allergic responses. Secretions can be collected directly, using absorptive materials placed on the mucosa [5] or by lavage with saline [6]. Cytokine levels can then be quantified by ELISA or other immunoassays. Lavage has the advantage of allowing concurrent cytological investigation, but the disadvantage of dilution of nasal fluid, which may render some mediators undetectable [7].

Nasal mucosal brushing or scraping can provide mRNA, allowing assessment of cytokine gene expression by real-time PCR [8]. Nasal biopsy allows use of immunohistochemistry and in situ hybridisation to quantify and localise cytokine-producing cells [9]. Nasal epithelial cell lines, obtained at

This article is part of the Topical Collection on *Rhinitis*

G. Scadding (✉)
Allergy and Clinical Immunology, Imperial College, London, South Kensington Campus, London SW7 2AZ, UK
e-mail: g.scadding@imperial.ac.uk

brushing or biopsy, may be investigated for cytokine production in vitro [10].

Cytokine production by isolated peripheral blood mononuclear cells, T cells and dendritic cells exposed to allergen in vitro has also been investigated in allergic rhinitis [11, 12•]. Serum levels of cytokines primarily produced in the nasal mucosa might be expected to be very low, but differences between allergic rhinitics and controls have been identified [13].

The timing of investigation is important. Studies of seasonal allergic rhinitis may be done both in and out of season, allowing comparison [14]. Given that allergen exposure in daily life is difficult to control for, an alternative approach is to use nasal allergen provocation. With this approach, applied doses can be standardised and the precise time course of response studied. An example of symptom and peak nasal inspiratory flow responses to nasal allergen challenge is shown in Fig. 1. A drawback to this approach is that it is markedly different in both timing and magnitude to allergen exposure in daily life. Conversely, environmental exposure chambers can provide controlled allergen exposure at commonly encountered levels over time [15].

It is also important to consider the profile of the patient group studied, such as perennial or seasonal rhinitics, the presence or absence of concurrent asthma, and the triggering allergen(s). Also, mono- versus polysensitisation may alter responses to a single allergen [16]. Lastly, the intrinsic protease activity of some allergens may influence innate immune responses in a non-IgE-dependent manner [17].

Th2 Cytokine Profiles

Nasal allergen provocation induces early phase symptoms – itching and sneezing, followed by rhinorrhoea and nasal blockage – within minutes of allergen exposure, accompanied by release of mast cell-derived mediators, including histamine [18], tryptase [6], PGD2 [18] and cysteinyl leukotrienes [19]. Neuropeptides are also elevated, including substance P, calcitonin gene-related peptide (CGRP) and vasoactive intestinal polypeptide (VIP) [20]. During the early phase response, however, Th2 cytokines remain at pre-challenge levels [5]. Whilst only a minority of patients show a distinct late phase clinical response (5; Scadding GW, Durham SR, unpublished data), an increase in IL-4, -5 and -13 may be detected in nasal fluid from 3-4 h onwards, rising to 6-9 h post challenge [5, 21-23]. Whether levels have reached a plateau by this point is unclear; by 24 h they are returning to baseline [24]. Levels of tryptase, IL-4, -5 and -13 in fluid directly absorbed from the nasal mucosa before and after grass pollen allergen challenge are shown in Fig. 2.

In high Th2-cytokine-secreting individuals, the correlation between different cytokines, particularly between IL-13 and IL-5, is strong, as illustrated in Fig. 3 [5]. Interleukin-5 appears to be present in highest concentration, with levels in excess of 1,000 pg/ml recorded, followed by IL-13 [5, 23, 25•]. Concentrations of IL-4 and IL-9 are lower, with a range of approximately 0-200 pg/ml for IL-4 and 0-100 pg/ml for IL-9 (5; Scadding, Durham unpublished data). The regulatory cytokine IL-10 is also present in nasal fluid and may be increased after nasal allergen challenge [26]. An increase in Th2 chemokines has also been reported. Eotaxin has been most frequently studied; increases in RANTES, MCP-1 and MIP-1α have also been identified [21, 24, 27, 28].

Studies of cytokine profiles during natural exposure to seasonal or perennial allergens provide information on the usual disease state of rhinitic patients, but not on the immediate time course of the allergic response. Increased levels of IL-4, IL-5 and IL-10, accompanied by elevated eosinophils, were found in nasal lavage fluid in school children with grass or tree pollen seasonal allergic rhinitis [29]. Direct absorption of nasal fluid with synthetic filter strips demonstrated elevated

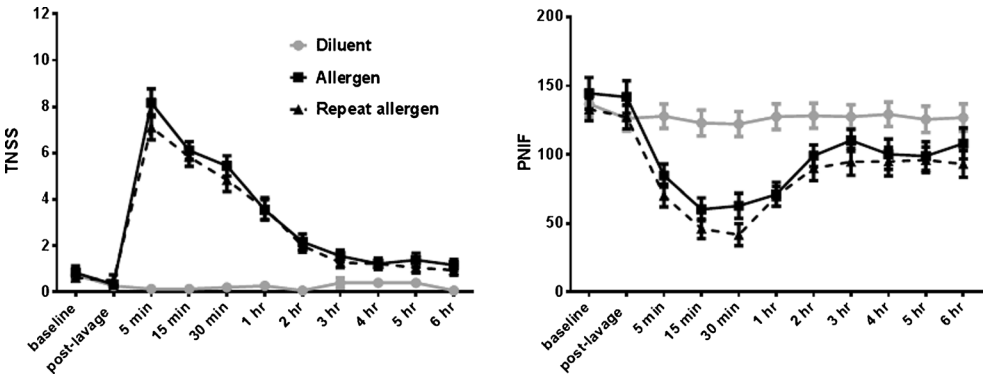

Fig. 1 Response to grass pollen nasal allergen challenge in allergic individuals. Purified Timothy grass (Phleum pratense) pollen allergen or diluent only was applied by nasal spray to both sides of the nasal mucosa at time 0, after initial assessments at baseline and following a nasal saline lavage. A repeat challenge with the same allergen dose was repeated approximately 4 weeks after the initial challenge. Symptom scores (TNSS, total nasal symptom score, 0-12) and peak nasal inspiratory flow (PNIF, L/min) were recorded before and after challenge for 6 h. Mean± SE of 18 volunteers

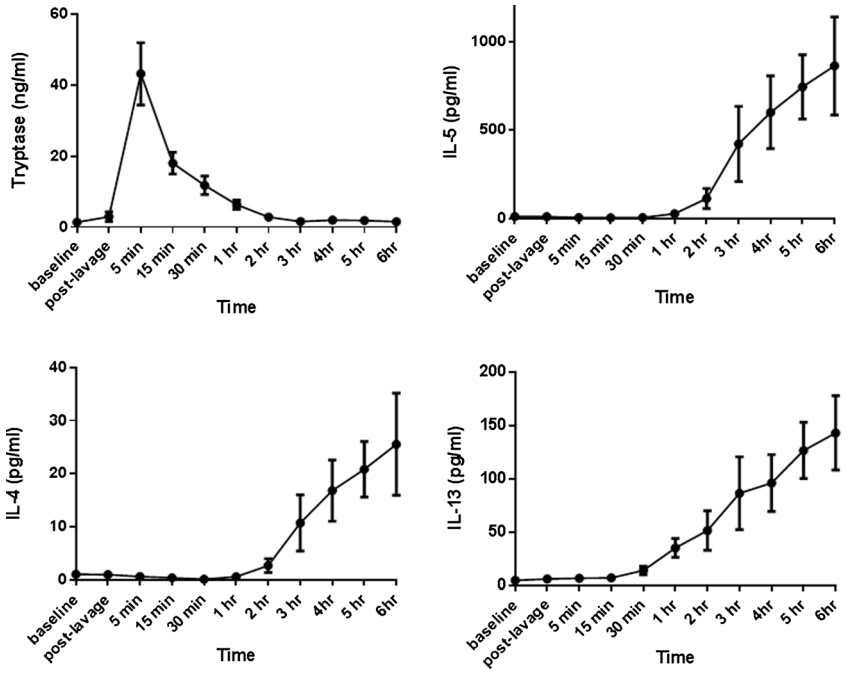

Fig. 2 Concentration of tryptase, IL-5, IL-4 and IL-13, measured by ImmunoCAP (tryptase; Phadia/Thermofisher) and MSD Human TH1/TH2 7-Plex (IL-5, IL-4, IL-13; MS6000 7 spot, Meso Scale Discovery) in nasal fluid absorbed directly from the nasal mucosa using polyurethane sponges and synthetic filter papers. Mean±SE of 18 volunteers. (*Adapted from* Scadding et al. [5] and unpublished data)

levels of IL-5, IL-13, eotaxin-1, TARC, MCP-1, MIP-1β and IP-10 in children with symptomatic allergic rhinitis [30]. Klemens et al. compared nasal fluid mediators in seasonal allergic rhinitis and viral rhinitis [31]. Allergics had elevated ECP, tryptase and IL-5; viral infection produced increased levels of a range of inflammatory cytokines, including IL-1β, IL-6, IL-7, IL-17, IFNγ, IL-8, TNFα and GM-CSF, but also IL-4 and IL-5. Interestingly, lower in-season IL-4 and IL-10 levels in nasal fluid have been reported in one study, but both were elevated at 5 h after nasal allergen challenge out of season, highlighting the potential discrepancies between the two approaches [32].

In situ hybridisation revealed elevated levels of IL-4 and IL-5 mRNA in nasal turbinate mucosal biopsies taken either in season [14, 33, 34] or 6 h after grass pollen allergen challenge [9]. Raised IL-10, IL-13 and RANTES mRNA post challenge have also been described [35]. A trend towards an increase in IL-9 mRNA in season was accompanied by an increase in c-kit+mast cells [36].

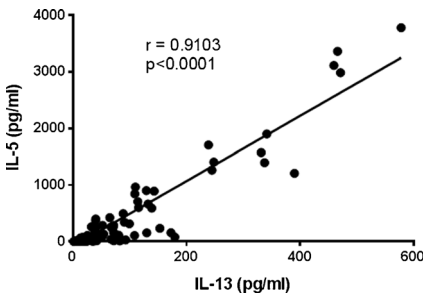

Fig. 3 Spearman correlation between IL-5 and IL-13 levels in nasal fluid of 18 patients before and after grass pollen nasal allergen challenge. (*Adapted from* Scadding et al. [5])

Immunohistochemical staining revealed fewer IL-10+ cells in the nasal mucosa of allergics after grass pollen challenge compared to controls (12). Peripheral blood myeloid dendritic cells from the same patients secreted less IL-10 in vitro and tended to support differentiation of naïve T cells into Th2 or Th17 cells rather than Th1 cells. This reveals a systemic discrepancy between allergics and non-atopics. In the same vein, a number of studies have reported elevated serum Th2 cytokines in allergics, including IL-5 [37] and IL-9 [38].

Several investigators have looked for associations between single nucleotide polymorphisms in Th2 cytokine genes and risk of allergic rhinitis, with IL-4 and IL-13 SNPs most frequently studied. Modest associations have been identified for some SNPs [39, 40] but not others [41]. Associations are likely to be population-specific, and may also be gender-specific [42].

In summary, both nasal allergen challenge and in-season studies demonstrate a clear Th2 cytokine profile in allergic rhinitis, with a time course in keeping with the classical model of late phase allergic inflammation. Further research is needed to determine how closely these profiles relate to clinical outcomes, the relationship between early phase mediators, such as tryptase, and late phase Th2 cytokines, and whether cytokine profiles can be used to predict response to treatment, particularly specific-allergen immunotherapy. The standardisation of procedures, particularly for allergen challenge and nasal fluid analysis, plus the use of increasingly sensitive, low-volume immunoassays, should enable more reliable comparison between studies and insight into these areas.

Th1 and Inflammatory Cytokines

Investigators have examined IFNγ as a measure of Th1 responses in allergic rhinitis. Most studies of symptomatic rhinitics have not identified increases in nasal fluid compared to controls or out-of-season levels [29, 31, 43]. In fact, in a study by Benson and colleagues, an increase in IFNγ during seasonal grass pollen exposure was seen in non-atopics, but not allergics. In this study, allergic status was best characterised by a high IL-4:IFNγ ratio in nasal fluid [29]. Similarly, lower tissue expression of IFNγ in turbinate biopsies from grass pollen allergics than controls was seen both before and after nasal challenge [12•]. Furthermore, in vitro, allergic peripheral blood plasmacytoid and myeloid dendritic cells released less IFNγ and IL-12, respectively [12•]. No significant increase was seen in IFNγ levels in nasal fluid following grass pollen challenge, despite increases in several other mediators [5].

Elevated nasal fluid IL-18 has been identified both during seasonal exposure in pollen allergics and, to a greater extent, in symptomatic dust mite allergics [44]. Conversely, elevated IL-1β levels were only found in seasonal rhinitics, in agreement with previous studies [45, 46]. Nasal allergen challenge has resulted in increased IL-1β, GM-CSF, IL-6 [27] and IL-8 [24] compared to baseline values. However, Klemens et al. [31] found no significant increases in IL-1β, IL-6, IL-8, GM-CSF, TNFα or the chemokines MCP1 and MIP-1β in the nasal fluid of seasonal allergic rhinitics, but did identify increases during viral upper respiratory tract infection.

Th17 Family Cytokines

The Th17 family of cytokines, particularly IL-17A (often referred to simply as IL-17), have proinflammatory effects in chronic asthma [47] and chronic rhinosinusitis—particularly chronic rhinosinusitis with nasal polyps in patients of Chinese ethnicity [48]. Their role in allergic rhinitis is less well established.

In a study of seasonal allergic rhinitics, IL-17A levels in nasal fluid increased 5 h after nasal allergen challenge, falling back to pre-challenge levels at 24 h [32]. However, during natural seasonal exposure in the same patients, IL-17A was largely undetectable. (It should be noted that different nasal fluid collection methods were used for the challenge and in-season phases of the study, potentially accounting for this difference.) Conversely, Xu et al. [10] did record elevated levels of IL-17A and IL-17 F (as well as IL-25/IL-17E and TSLP) by nasal lavage in unchallenged house dust mite allergics. This raises the possibility that cytokine profiles differ between pollen- and house dust mite-induced rhinitis (perhaps as a result of intrinsic Der p1 protease activity), although methodological differences may again be relevant. Additionally, the populations studied—European and Chinese, respectively—might also account for differences, particularly in light of the different cytokine profiles described in European and Chinese nasal polyps [48]. Whilst IL-17 was found to be increased in nasal fluid in a European population during viral infection, no increase was found during seasonal allergen exposure [31]. Nasal allergen challenge with grass pollen did not produce increases in nasal fluid IL-17 despite increases in several Th2 cytokines and chemokines [49].

Two studies have reported increased serum IL-17A levels in allergic rhinitics: during the pollen season [50], and before and after house dust mite bronchial challenge [51]. In the latter study, the concentration was intermediate between non-atopic controls and allergic asthmatics.

Inferior turbinate tissue from perennial dust mite allergic rhinitics had greater IL-17A+cells by immunohistochemistry and IL-17A mRNA by real-time PCR, and an increased proportion of CD4+IL-17A+cells in tissue homogenates compared to non-allergic controls [52]. But IL-17A expression did not correlate with clinical symptoms.

Peripheral blood myeloid dendritic cells isolated from grass pollen allergics had increased propensity to induce T cell IL-17 secretion in vitro [12•]. Grass or birch pollen allergen-challenged peripheral blood mononuclear cells were found to have significantly upregulated IL-17 receptor (IL-17RB) gene expression – more so than IL-5, GATA-3 or FceRII. Protein levels of IL-17RB were also increased on basophils post challenge [53].

Overall, evidence suggests that IL-17 may be elevated in allergic rhinitis, predominantly in mite-induced, perennial rhinitis. Whether it has a functional, pathological role in allergic rhinitis is unclear. Areas of potential research interest include treatment effects on IL-17, influence of IL-17 on response to treatment, particularly to intra-nasal corticosteroids, and whether elevated IL-17 may put individual rhinitics at increased risk of developing asthma or rhinosinusitis.

Epithelial-derived Cytokines

Recent evidence from murine asthma models suggests that airways inflammation and hyper-reactivity are dependent on cytokines secreted predominantly by the airway epithelium, including TSLP, IL-33 and IL-25. These cytokines are released by tissue damage, pathogen recognition or even by allergen exposure. They affect Th2 cell function either directly or via innate lymphoid cells, which in turn produce IL-5, IL-9 and IL-13 [54]. These novel cytokines and innate lymphoid cells appear to be relevant to both human asthma [55, 56] and chronic rhinosinusitis with nasal polyps [57•, 58••]; their relevance in allergic rhinitis is also under investigation.

A higher concentration of TSLP was found in nasal lavage fluid from unchallenged house dust mite sensitised allergic rhinitics than non-atopic controls [10], at a mean of 33.8 pg/ml in allergics. In the same study, dsRNA induced TSLP release from human nasal epithelial cells in vitro. Immunohistochemical staining and real-time PCR have revealed increased expression of TSLP protein and mRNA in turbinate tissue of allergic rhinitics [59, 60]. TSLP production by human nasal epithelial cells in vitro was stimulated by a TLR2 ligand, as well as by IL-1β and TNFα [59]. A further study identified increased TSLP expression in vitro in human nasal epithelial cells derived from mugwort allergics in season compared to controls [61]. The results of these interesting studies require confirmation in larger, well-characterised cohorts. Of note, in a study of 11 TSLP single-nucleotide polymorphisms in a Han Chinese population, none were associated with susceptibility to allergic rhinitis [62]; but the same SNPs had gender-specific associations with nasal polyposis [63].

Data concerning IL-33 are conflicting. Levels of approximately 5 ng/ml were found in nasal secretions (collected by aspiration) of a cohort of house dust mite and Japanese cedar allergic rhinitics – significantly higher than in non-atopic controls [64] – but IL-33 was undetectable in serum in either group. Conversely, IL-33 was undetectable in nasal lavage fluid collected during seasonal pollen exposure and present at only low pg/ml levels, without a significant increase following nasal challenge using direct mucosal fluid absorption with filter discs [65]. In this latter study, however, the soluble IL-33 receptor, ST2, was elevated in the nasal fluid of allergics in season. Serum IL-33 has been detected by other researchers, at varying levels: a median of 549 pg/ml in Japanese cedar allergics [13], 2133 pg/ml in house dust mite allergics [66] and 28.5 ng/ml in grass and tree allergics [67]. In each of these studies, the serum concentration was greater than in non-atopic controls.

Haenuki et al. reported reduced turbinate epithelial expression of IL-33 on immunohistochemical staining in allergic rhinitics. However, IL-33 mRNA expression was increased in pollen allergics biopsied in season [68•]. Conversely, increased IL-33 protein and mRNA expression has been reported in biopsy tissue from house dust mite allergics [66]. In this latter study, human nasal epithelial cell IL-33 expression in vitro was induced by either IFNγ or a TLR9 ligand. Finally, a weak association between an IL-33 gene single nucleotide polymorphism and Japanese cedar pollinosis has been reported [13].

A convincing role for epithelial-derived cytokines in allergic rhinitis in man has yet to be proven, although this area is now of great research interest. As in asthma, these possible upstream inflammatory mediators would make very attractive targets for pharmacotherapy, with the potential for more diverse inhibitory effects than is seen with targeting individual Th2 cytokines.

Other Cytokines and Mediators

Periostin [69], osteopontin [37] and IL-31 [70-72] have all been studied in the context of allergic rhinitis, with greater levels found than in controls, thus indicating possible pathological roles. Conversely, the anti-inflammatory protein CC10 (Clara Cell 10kD protein) has been identified in reduced levels in allergic rhinitic nasal mucosa, with an inverse correlation with osteopontin expression [73].

Treatment Effects on Cytokine Profiles

Intranasal corticosteroids are the most effective pharmacotherapy for allergic rhinitis. They decrease eosinophil infiltration into the nasal mucosa during seasonal allergen exposure [33]. This may be due to their ability to suppress local mucosal IL-5, as detected in nasal fluid [34, 74], or at mRNA level by in situ hybridisation [33]. Intranasal corticosteroids also inhibit increases in IL-4, -5 and -13 after nasal allergen challenge [21, 24] and prevent seasonal increases in IL-4 [14] and eotaxin [75]. They appear to have a less pronounced effect on seasonal neutrophil infiltration into the nasal mucosa [74] and accompanying inflammatory or Th1 cytokines, including IFNγ, IL-1β and TNFα [76]. However, topical fluticasone did significantly inhibit IL-1β, IL-8, IL-6 and MIP-1α release in nasal fluid following nasal allergen challenge [77].

Specific allergen immunotherapy is highly effective when used in appropriate patients and induces lasting immunological tolerance [78] associated with production of functional, allergen-specific IgG4 antibodies [79]. Grass pollen immunotherapy has been shown to inhibit the seasonal rise in eosinophil infiltration into the nasal mucosa as well as reduce IL-5 mRNA expression in tissue [80] and IL-5 protein in nasal fluid [81]. Interferon-γ mRNA levels are increased, with clinical improvement accompanied by an increase in the local IFNγ:IL-5 ratio [82]. Local IL-10 mRNA expression is increased, as is IL-10 production by peripheral blood T cells in vitro [83]. There is a reduction in IL-9 alongside reduced c-kit+mast cells [36]. Immunotherapy may also inhibit allergen-induced peripheral blood T cell IL-4 production in vitro [84].

Conclusions

Allergic rhinitis is associated with a dominant Th2 cytokine profile, which is suppressed by corticosteroids. The profiles seen following either nasal allergen challenge or in-season/symptomatic assessment are broadly similar, although the magnitude of cytokines may be greater following high dose challenge. Controlled allergen challenge has the advantage of providing a time course of mediator release, demonstrating that Th2 cytokines increase from basal levels as early as 3-4 h

post challenge, reaching peak levels at or after 6-8 h and returning to near baseline levels by 24 h. The precise cellular source of these cytokines requires further clarification. Whilst infiltrating T cells and eosinophils are likely to contribute, the rapid appearance of these cytokines in nasal fluid suggests local resident cells may also be involved.

Innate lymphoid cells have been recognised as a source of Th2 cytokines, particularly IL-13, in mouse models of allergic asthma. A role for these cells in human allergic rhinitis has to be established, but research on the epithelial-derived cytokines believed to be responsible for activating innate lymphoid cells is now taking off and beginning to provide interesting results. Further investigation is required.

With increasing realisation of the interaction between upper and lower airways, it is important to look for common pathomechanisms. Emerging evidence in asthma suggests it is a heterogeneous disease, with some forms being less corticosteroid responsive. This pattern may, in part, be accounted for by IL-17/Th17-cell-mediated, neutrophilic inflammation. Predominant Th17-type inflammation has also been demonstrated in some patients with chronic rhinosinusitis with nasal polyps. A clear role for IL-17 family cytokines in allergic rhinitis has yet to be confirmed, with results at present conflicting. As is the emerging trend with asthma, it may one day be possible to differentiate allergic rhinitics into various phenotypes (including differential corticosteroid-responsiveness), based in part on cytokine profiles, allowing for improved, targeted treatments.

Compliance with Ethics Guidelines

Conflict of Interest Guy Scadding is currently an Imperial College Wellcome Trust Clinical PhD Fellow, with funding provided by The Wellcome Trust through Imperial College (from October 2011 to October 2014).

Human and Animal Rights and Informed Consent This article does not contain any studies with animal subjects performed by the author. With regard to the author's research cited in this paper, all procedures were followed in accordance with the ethical standards of the responsible committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 2000 and 2008.

References

Papers of particular interest, published recently, have been highlighted as:

- • Of importance
- •• Of major importance

1. Maes T, Joos GF, Brusselle GG. Targeting interleukin-4 in asthma: lost in translation? Am J Respir Cell Mol Biol. 2012;47(3):261–70.
2. Cosmi L, Liotta F, Maggi E, Romagnani S, Annunziato F. Th17 cells: new players in asthma pathogenesis. Allergy. 2011;66(8): 989–98.
3. Ying S, O'Connor B, Ratoff J, Meng Q, Fang C, Cousins D, et al. Expression and cellular provenance of thymic stromal lymphopoietin and chemokines in patients with severe asthma and chronic obstructive pulmonary disease. J Immunol. 2008;181(4):2790–8.
4. Lloyd CM. IL-33 family members and asthma – bridging innate and adaptive immune responses. Curr Opin Immunol. 2010;22(6): 800–6.
5. Scadding GW, Calderon MA, Bellido V, Koed GK, Nielsen NC, Lund K, et al. Optimisation of grass pollen nasal allergen challenge for assessment of clinical and immunological outcomes. J Immunol Methods. 2012;384(1–2):25–32.
6. Castells M, Schwartz LB. Tryptase levels in nasal-lavage fluid as an indicator of the immediate allergic response. J Allergy Clin Immunol. 1988;82(3 Pt 1):348–55.
7. Riechelmann H, Deutschle T, Friemel E, et al. Biological markers in nasal secretions. Eur Respir J. 2003;21(4):600–5.
8. Kitamura Y, Mizuguchi H, Ogishi H, Kuroda W, Hattori M, Fukui H, et al. Preseasonal prophylactic treatment with antihistamines suppresses IL-5 but not IL-33 mRNA expression in the nasal mucosa of patients with seasonal allergic rhinitis caused by Japanese cedar pollen. Acta Otolaryngol. 2012;132(4):434–8.
9. Nouri-Aria KT, O'Brien F, Noble W, et al. Cytokine expression during allergen-induced late nasal responses: IL-4 and IL-5 mRNA is expressed early (at 6 h) predominantly by eosinophils. Clin Exp Allergy. 2000;30(12):1709–16.
10. Xu G, Zhang L, Wang DY, Xu R, Liu Z, Han DM, et al. Opposing roles of IL-17A and IL-25 in the regulation of TSLP production in human nasal epithelial cells. Allergy. 2010;65(5):581–9.
11. Till S, Durham S, Dickason R, Huston D, Bungre J, Walker S, et al. IL-13 production by allergen-stimulated T cells is increased in allergic disease and associated with IL-5 but not IFN-gamma expression. Immunology. 1997;91(1):53–7.
12. • Pilette C, Jacobson MR, Ratajczak C, Detry B, Banfield G, VanSnick J, et al. Aberrant dendritic cell function conditions Th2-cell polarization in allergic rhinitis. Allergy. 2013;68(3):312–21. *Alterations in dendritic cell function in allergic rhinitis demonstrated both locally in the nasal mucosa and in the peripheral circulation.*
13. Sakashita M, Yoshimoto T, Hirota T, Harada M, Okubo K, Osawa Y, et al. Association of serum interleukin-33 level and the interleukin-33 genetic variant with Japanese cedar pollinosis. Clin Exp Allergy. 2008;38(12):1875–81.
14. Cameron LA, Durham SR, Jacobson MR, Masuyama K, Juliusson S, Gould HJ, et al. Expression of IL-4, Cepsilon RNA, and Iepsilon RNA in the nasal mucosa of patients with seasonal rhinitis: effect of topical corticosteroids. J Allergy Clin Immunol. 1998;101(3):330–6.
15. Patel D, Couroux P, Hickey P, Salapatek AM, Laidler P, Larché M, Hafner RP. Fel d 1-derived peptide antigen desensitization shows a persistent treatment effect 1 year after the start of dosing: a randomized, placebo-controlled study. J Allergy Clin Immunol. 2013;131(1):103-9.e1-7
16. Reinartz SM, van Ree R, Versteeg SA, Zuidmeer L, van Drunen CM, Fokkens WJ. Diminished response to grass pollen allergen challenge in subjects with concurrent house dust mite allergy. Rhinology. 2009;47(2):192–8.
17. Kikuchi Y, Takai T, Kuhara T, Ota M, Kato T, Hatanaka H, et al. Crucial commitment of proteolytic activity of a purified recombinant major house dust mite allergen Der p1 to sensitization toward IgE and IgG responses. J Immunol. 2006;177(3):1609–17.
18. Naclerio RM, Proud D, Togias AG, Adkinson Jr NF, Meyers DA, Kagey-Sobotka A, et al. Inflammatory mediators in late antigen-induced rhinitis. N Engl J Med. 1985;313(2):65–70.
19. Creticos PS, Peters SP, Adkinson Jr NF, Naclerio RM, Hayes EC, Norman PS, et al. Peptide leukotriene release after antigen

challenge in patients sensitive to ragweed. N Engl J Med. 1984;310(25):1626–30.
20. Mosimann BL, White MV, Hohman RJ, Goldrich MS, Kaulbach HC, Kaliner MA. Substance P, calcitonin gene-related peptide, and vasoactive intestinal peptide increase in nasal secretions after allergen challenge in atopic patients. J Allergy Clin Immunol. 1993;92(1 Pt 1):95–104.
21. Erin EM, Zacharasiewicz AS, Nicholson GC, Tan AJ, Higgins LA, Williams TJ, et al. Topical corticosteroid inhibits interleukin-4, -5 and -13 in nasal secretions following allergen challenge. Clin Exp Allergy. 2005;35(12):1608–14.
22. Wagenmann M, Schumacher L, Bachert C. The time course of the bilateral release of cytokines and mediators after unilateral nasal allergen challenge. Allergy. 2005;60(9):1132–8.
23. Linden M, Svensson C, Andersson E, Andersson M, Greiff L, Persson CG. Immediate effect of topical budesonide on allergen challenge-induced nasal mucosal fluid levels of granulocyte-macrophage colony-stimulating factor and interleukin-5. Am J Respir Crit Care Med. 2000;162(5):1705–8.
24. Erin EM, Leaker BR, Zacharasiewicz AS, et al. Single dose topical corticosteroid inhibits IL-5 and IL-13 in nasal lavage following grass pollen challenge. Allergy. 2005;60(12):1524–9.
25.• Nicholson GC, Kariyawasam HH, Tan AJ, et al. The effects of an anti-inflammatory IL-13 mAb on cytokine levels and nasal symptoms following nasal allergen challenge. J Allergy Clin Immunol. 2011;128(4):800–7. *Systemic administration of anti-IL-13 blocking antibody reduces nasal fluid IL-13 after nasal allergen challenge.*
26. Bensch GW, Nelson HS, Borish LC. Evaluation of cytokines in nasal secretions after nasal antigen challenge: lack of influence of antihistamines. Ann Allergy Asthma Immunol. 2002;88(5):457–62.
27. Sim TC, Reece LM, Hilsmeier KA, Grant JA, Alam R. Secretion of chemokines and other cytokines in allergen-induced nasal responses: inhibition by topical steroid treatment. Am J Respir Crit Care Med. 1995;152(3):927–33.
28. Terada N, Hamano N, Kim WJ, Hirai K, Nakajima T, Yamada H, et al. The kinetics of allergen-induced eotaxin level in nasal lavage fluid: its key role in eosinophil recruitment in nasal mucosa. Am J Respir Crit Care Med. 2001;164(4):575–9.
29. Benson M, Strannegård IL, Wennergren G, Strannegård O. Cytokines in nasal fluids from school children with seasonal allergic rhinitis. Pediatr Allergy Immunol. 1997;8(3):143–9.
30. Chawes BL, Edwards MJ, Shamji B, Walker C, Nicholson GC, Tan AJ, et al. A novel method for assessing unchallenged levels of mediators in nasal epithelial lining fluid. J Allergy Clin Immunol. 2010;125(6):1387–9.
31. Klemens C, Rasp G, Jund F, Hilgert E, Devens C, Pfrogner E. Kramer MF Mediators and cytokines in allergic and viral-triggered rhinitis. Allergy Asthma Proc. 2007;28(4):434–41.
32. Baumann R, Rabaszowski M, Stenin I, Tilgner L, Scheckenbach K, Wiltfang J, et al. Comparison of the nasal release of IL-4, IL-10, IL-17, CCL13/MCP-4, and CCL26/eotaxin-3 in allergic rhinitis during season and after allergen challenge. Am J Rhinol Allergy. 2013;27(4):266–72.
33. Masuyama K, Till SJ, Jacobson MR, Kamil A, Cameron L, Juliusson S, et al. Nasal eosinophilia and IL-5 mRNA expression in seasonal allergic rhinitis induced by natural allergen exposure: effect of topical corticosteroids. J Allergy Clin Immunol. 1998;102(4 Pt 1):610–7.
34. Kita H, Jorgensen RK, Reed CE, Dunnette SL, Swanson MC, Bartemes KR, et al. Mechanism of topical glucocorticoid treatment of hay fever: IL-5 and eosinophil activation during natural allergen exposure are suppressed, but IL-4, IL-6, and IgE antibody production are unaffected. J Allergy Clin Immunol. 2000;106(3):521–9.
35. KleinJan A, Dijkstra MD, Boks SS, Severijnen LA, Mulder PG, Fokkens WJ. Increase in IL-8, IL-10, IL-13, and RANTES mRNA levels (in situ hybridization) in the nasal mucosa after nasal allergen provocation. J Allergy Clin Immunol. 1999;103(3 Pt 1):441–50.
36. Nouri-Aria KT, Pilette C, Jacobson MR, Watanabe H, Durham SR. IL-9 and c-Kit+mast cells in allergic rhinitis during seasonal allergen exposure: effect of immunotherapy. J Allergy Clin Immunol. 2005;116(1):73–9.
37. Liu W, Xia W, Fan Y, Wang H, Zuo K, Lai Y, et al. Elevated serum osteopontin level is associated with blood eosinophilia and asthma comorbidity in patients with allergic rhinitis. J Allergy Clin Immunol. 2012;130(6):1416–8.
38. Ciprandi G. Serum interleukin 9 in allergic rhinitis. Ann Allergy Asthma Immunol. 2010;104(2):180–1.
39. Ying XJ, Zhao SW, Wang GL, Xie J, Xu HM, Dong P. Association of interleukin-13 SNP rs20541 with allergic rhinitis risk: a meta-analysis. Gene. 2013;521(2):222–6.
40. Movahedi M, Amirzargar AA, Nasiri R, Hirbod-Mobarakeh A, Farhadi E, Tavakol M, et al. Gene polymorphisms of Interleukin-4 in allergic rhinitis and its association with clinical phenotypes. Am J Otolaryngol. 2013;34(6):676–81.
41. Ying X, Zhang R, Yu S, Wu J, Wang H. Association of interleukin-13 SNP rs1800925 with allergic rhinitis risk: a meta-analysis based on 1,411 cases and 3169 controls. Gene. 2012;506(1):179–83.
42. Miyake Y, Tanaka K, Arakawa M. Polymorphisms in the IL4 gene, smoking, and rhinoconjunctivitis in Japanese women: the Kyushu Okinawa Maternal and Child Health Study. Hum Immunol. 2012;73(10):1046–9.
43. Gröger M, Klemens C, Wendt S, Becker S, Canis M, Havel M, et al. Mediators and cytokines in persistent allergic rhinitis and nonallergic rhinitis with eosinophilia syndrome. Int Arch Allergy Immunol. 2012;159(2):171–8.
44. Verhaeghe B, Gevaert P, Holtappels G, Lukat KF, Lange B, Van Cauwenberge P, et al. Up-regulation of IL-18 in allergic rhinitis. Allergy. 2002;57(9):825–30.
45. Bachert C, van Kempen M, Van Cauwenberge P. Regulation of proinflammatory cytokines in seasonal allergic rhinitis. Int Arch Allergy Immunol. 1999;118(2–4):375–9.
46. Sim TC, Grant JA, Hilsmeier KA, Fukuda Y, Alam R. Proinflammatory cytokines in nasal secretions of allergic subjects after antigen challenge. Am J Respir Crit Care Med. 1994;149(2 Pt 1):339–44.
47. Molet S, Hamid Q, Davoine F, Nutku E, Taha R, Pagé N, et al. IL-17 is increased in asthmatic airways and induces human bronchial fibroblasts to produce cytokines. J Allergy Clin Immunol. 2001;108(3):430–8.
48. Zhang N, Van Zele T, Perez-Novo C, Van Bruaene N, Holtappels G, DeRuyck N, et al. Different types of T-effector cells orchestrate mucosal inflammation in chronic sinus disease. J Allergy Clin Immunol. 2008;122(5):961–8.
49. Scadding GW, Eifan A, Penagos M, Koed GK, Shamji MH, Wurtzen PA, Durham SR. Grass pollen nasal challenge is associated with increases in Th2 cytokines, Eotaxin, MDC and IL-6 in nasal fluid; Th1 and Th17-associated cytokines are low and do not increase following challenge. Abstract; EAACI SERIN meeting, Leuven, March 2013.
50. Ciprandi G, Fenoglio D, De Amici M, Quaglini S, Negrini S, Filaci G. Serum IL-17 levels in patients with allergic rhinitis. J Allergy Clin Immunol. 2008;122(3):650–1.
51. Bajoriuniene I, Malakauskas K, Lavinskiene S, Jeroch J, Gasiuniene E, Vitkauskiene A, et al. Response of peripheral blood Th17 cells to inhaled Dermatophagoides pteronyssinus in patients with allergic rhinitis and asthma. Lung. 2012;190(5):487–95.
52. Liu Y, Yu HJ, Wang N, Zhang YN, Huang SK, Cui YH, et al. Clara cell 10-kDa protein inhibits T(H)17 responses through modulating dendritic cells in the setting of allergic rhinitis. J Allergy Clin Immunol. 2013;131(2):387–94.

53. Wang H, Mobini R, Fang Y, Barrenäs F, Zhang H, Xiang Z, et al. Allergen challenge of peripheral blood mononuclear cells from patients with seasonal allergic rhinitis increases IL-17RB, which regulates basophil apoptosis and degranulation. Clin Exp Allergy. 2010;40(8):1194–202.
54. Licona-Limón P, Kim LK, Palm NW, Flavell RA. TH2, allergy and group 2 innate lymphoid cells. Nat Immunol. 2013;14(6):536–42.
55. Saglani S, Lui S, Ullmann N, Campbell GA, Sherburn RT, Mathie SA, et al. IL-33 promotes airway remodeling in pediatric patients with severe steroid-resistant asthma. J Allergy Clin Immunol. 2013;132(3):676–85.
56. Shikotra A, Choy DF, Ohri CM, Doran E, Butler C, Hargadon B, et al. Increased expression of immunoreactive thymic stromal lymphopoietin in patients with severe asthma. J Allergy Clin Immunol. 2012;129(1):104–11.
57.• Shaw JL, Fakhri S, Citardi MJ, Porter PC, Corry DB, Kheradmand F, et al. IL-33-responsive innate lymphoid cells are an important source of IL-13 in chronic rhinosinusitis with nasal polyps. Am J Respir Crit Care Med. 2013;188(4):432–9. *Elevated levels of ST2+ innate lymphoid cells in ethmoid sinus mucosa in patients with chronic rhinosinusitis with nasal polyps compared to patients with chronic rhinosinusitis without nasal polyps or healthy controls.*
58.•• Mjösberg JM, Trifari S, Crellin NK, Peters CP, van Drunen CM, Piet B, et al. Human IL-25- and IL-33-responsive type 2 innate lymphoid cells are defined by expression of CRTH2 and CD161. Nat Immunol. 2011;12(11):1055–62. *Description of a CRTH2+ innate lymphoid cell population in humans with enrichment in nasal polyp tissue.*
59. Kamekura R, Kojima T, Koizumi J, Ogasawara N, Kurose M, Go M, et al. Thymic stromal lymphopoietin enhances tight-junction barrier function of human nasal epithelial cells. Cell Tissue Res. 2009;338(2):283–93.
60. Mou Z, Xia J, Tan Y, Wang X, Zhang Y, Zhou B, et al. Overexpression of thymic stromal lymphopoietin in allergic rhinitis. Acta Otolaryngol. 2009;129(3):297–301.
61. Zhu DD, Zhu XW, Jiang XD, Dong Z. Thymic stromal lymphopoietin expression is increased in nasal epithelial cells of patients with mugwort pollen sensitive-seasonal allergic rhinitis. Chin Med J (Engl). 2009;122(19):2303–7.
62. Zhang Y, Song X, Zhao Y, Zhang L, Bachert C. Single nucleotide polymorphisms in thymic stromal lymphopoietin gene are not associated with allergic rhinitis susceptibility in Chinese subjects. BMC Med Genet. 2012;13:79.
63. Zhang Y, Wang X, Zhang W, Han D, Zhang L, Bachert C. Polymorphisms in thymic stromal lymphopoietin gene demonstrate a gender and nasal polyposis-dependent association with chronic rhinosinusitis. Hum Immunol. 2013;74(2):241–8.
64. Asaka D, Yoshikawa M, Nakayama T, Yoshimura T, Moriyama H, Otori N. Elevated levels of interleukin-33 in the nasal secretions of patients with allergic rhinitis. Int Arch Allergy Immunol. 2012;158 Suppl 1:47–50.
65. Baumann R, Rabaszowski M, Stenin I, Tilgner L, Gaertner-Akerboom M, Scheckenbach K, et al. Nasal levels of soluble IL-33R ST2 and IL-16 in allergic rhinitis: inverse correlation trends with disease severity. Clin Exp Allergy. 2013;43(10):1134–43.
66. Kamekura R, Kojima T, Takano K, Go M, Sawada N, Himi T. The role of IL-33 and its receptor ST2 in human nasal epithelium with allergic rhinitis. Clin Exp Allergy. 2012;42(2):218–28.
67. Glück J, Rymarczyk B, Rogala B. Serum IL-33 but not ST2 level is elevated in intermittent allergic rhinitis and is a marker of the disease severity. Inflamm Res. 2012;61(6):547–50.
68.• Haenuki Y, Matsushita K, Futatsugi-Yumikura S, Ishii KJ, Kawagoe T, Imoto Y, et al. A critical role of IL-33 in experimental allergic rhinitis. J Allergy Clin Immunol. 2012;130(1):184–94. *IL-33 required for manifestation of ragweed-induced allergic rhinitis in a mouse model; tissue expression of IL-33 protein reduced in the epithelium of allergic rhinitic patients compared to controls.*
69. Ishida A, Ohta N, Suzuki Y, Kakehata S, Okubo K, Ikeda H, et al. Expression of pendrin and periostin in allergic rhinitis and chronic rhinosinusitis. Allergol Int. 2012;61(4):589–95.
70. Baumann R, Rabaszowski M, Stenin I, Gaertner-Akerboom M, Scheckenbach K, Wiltfang J, et al. The release of IL-31 and IL-13 after nasal allergen challenge and their relation to nasal symptoms. Clin Transl Allergy. 2012;2(1):13.
71. Shah SA, Ishinaga H, Hou B, Okano M, Takeuchi K. Effects of interleukin-31 on MUC5AC gene expression in nasal allergic inflammation. Pharmacology. 2013;91(3–4):158–64.
72. Okano M, Fujiwara T, Higaki T, Makihara S, Haruna T, Noda Y, et al. Characterization of pollen antigen-induced IL-31 production by PBMCs in patients with allergic rhinitis. J Allergy Clin Immunol. 2011;127(1):277–9.
73. Wang H, Liu Y, Liu Z. Clara cell 10-kD protein in inflammatory upper airway diseases. Curr Opin Allergy Clin Immunol. 2013;13(1):25–30.
74. Benson M, Strannegård IL, Wennergren G, Strannegård O. Interleukin-5 and interleukin-8 in relation to eosinophils and neutrophils in nasal fluids from school children with seasonal allergic rhinitis. Pediatr Allergy Immunol. 1999;10(3):178–85.
75. Pullerits T, Lindén A, Praks L, Cardell LO, Lötvall J. Upregulation of nasal mucosal eotaxin in patients with allergic rhinitis during grass pollen season: effect of a local glucocorticoid. Clin Exp Allergy. 2000;30(10):1469–75.
76. Benson M, Strannegård IL, Strannegård O, Wennergren G. Topical steroid treatment of allergic rhinitis decreases nasal fluid TH2 cytokines, eosinophils, eosinophil cationic protein, and IgE but has no significant effect on IFN-gamma, IL-1beta, TNF-alpha, or neutrophils. J Allergy Clin Immunol. 2000;106(2):307–12.
77. Weido AJ, Reece LM, Alam R, Cook CK, Sim TC. Intranasal fluticasone propionate inhibits recovery of chemokines and other cytokines in nasal secretions in allergen-induced rhinitis. Ann Allergy Asthma Immunol. 1996;77(5):407–15.
78. Durham SR, Walker SM, Varga EM, Jacobson MR, O'Brien F, Noble W, et al. Long-term clinical efficacy of grass-pollen immunotherapy. N Engl J Med. 1999;341(7):468–75.
79. Shamji MH, Ljørring C, Francis JN, Calderon MA, Larché M, Kimber I, et al. Functional rather than immunoreactive levels of IgG4 correlate closely with clinical response to grass pollen immunotherapy. Allergy. 2012;67(2):217–26.
80. Wilson DR, Nouri-Aria KT, Walker SM, Pajno GB, O'Brien F, Jacobson MR, et al. Grass pollen immunotherapy: symptomatic improvement correlates with reductions in eosinophils and IL-5 mRNA expression in the nasal mucosa during the pollen season. J Allergy Clin Immunol. 2001;107(6):971–6.
81. Klimek L, Dormann D, Jarman ER, Cromwell O, Riechelmann H, Reske-Kunz AB. Short-term preseasonal birch pollen allergoid immunotherapy influences symptoms, specific nasal provocation and cytokine levels in nasal secretions, but not peripheral T-cell responses, in patients with allergic rhinitis. Clin Exp Allergy. 1999;29(10):1326–35.
82. Wachholz PA, Nouri-Aria KT, Wilson DR, Walker SM, Verhoef A, Till SJ, et al. Grass pollen immunotherapy for hayfever is associated with increases in local nasal but not peripheral Th1:Th2 cytokine ratios. Immunology. 2002;105(1):56–62.
83. Nouri-Aria KT, Wachholz PA, Francis JN, Jacobson MR, Walker SM, Wilcock LK, et al. Grass pollen immunotherapy induces mucosal and peripheral IL-10 responses and blocking IgG activity. J Immunol. 2004;172(5):3252–9.
84. Secrist H, Chelen CJ, Wen Y, Marshall JD, Umetsu DT. Allergen immunotherapy decreases interleukin 4 production in CD4+ T cells from allergic individuals. J Exp Med. 1993;178(6):2123–30.