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Rhinovirus and childhood asthma: an update

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Asthma is recognized as a complex disease resulting from interactions between multiple genetic and environmental factors. Accumulating evidence suggests that respiratory viral infections in early life constitute a major environmental risk factor for the development of childhood asthma. Respiratory viral infections have also been recognized as the most common cause of asthma exacerbation. The advent of molecular diagnostics to detect respiratory viruses has provided new insights into the role of human rhinovirus (HRV) infections in the pathogenesis of asthma. However, it is still unclear whether HRV infections cause asthma or if wheezing with HRV infection is simply a predictor of childhood asthma. Recent clinical and experimental studies have identified plausible pathways by which HRV infection could cause asthma, particularly in a susceptible host, and exacerbate disease. Airway epithelial cells, the primary site of infection and replication of HRV, play a key role in these processes. Details regarding the role of genetic factors, including *ORMDL3*, are beginning to emerge. This review discusses recent clinical and experimental evidence for the role of HRV infection in the development and exacerbation of childhood asthma and the potential underlying mechanisms that have been proposed.

Key words: Rhinovirus, Asthma, Epithelial cells, Cytokines

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Introduction

Asthma is recognized as a complex disease resulting from interactions between multiple genetic and environmental factors. Cumulative evidence suggests that respiratory viral infections in early life constitute a major environmental risk factor for the development of asthma[1-3]. Previously, most research in this field focused on respiratory syncytial virus (RSV) infection[4-8]. However, application of molecular diagnostic techniques to detect respiratory viruses has led to an increased awareness of the role of human rhinovirus (HRV) in wheezing illnesses during infancy and childhood asthma. Furthermore, recent large birth cohort studies have even suggested that HRV infection in the first years of life could be a much stronger risk factor for the subsequent development of asthma than RSV infection, at least in high-risk populations[9]. HRVs are also the most common viral pathogens isolated from children and adults with asthma exacerbations. Nonetheless, the fundamental mechanism that underlies these associations is still largely unknown. This review discusses the recent clinical and experimental evidence for the role of HRV infection in the development and exacerbation of childhood asthma and the potential underlying mechanisms that have been proposed. A greater understanding of the relationship between HRV infections, allergic inflammation, and asthma could lead to new strategies to prevent and treat childhood asthma.

HRV and host innate immune responses

HRVs, members of the family *Picornaviridae*, are positive-sense, single-stranded RNA viruses with a genome of approximately 7.2 kb. Until recently, HRVs have been classified into 2 species, HRV-A (containing 74 serotypes) and HRV-B (containing 25 serotypes), based on phylogenetic sequence criteria[10]. The development of highly sensitive molecular techniques to detect HRV in clinical specimens led to the identification and designation of a novel species, HRV-C. More than 60 types of this new species have since been recognized[11].

HRVs are also classified on the basis of receptor specificity. More than 90% of HRV serotypes, known as the "major group," utilize the intercellular adhesion molecule 1 (ICAM-1) as a receptor to enter host cells[12,13], whereas the "minor group" of HRVs utilizes the low-density lipoprotein receptor (LDLR). The major group consists of both HRV-A and HRV-B species, whereas all members of the minor group are HRV-A viruses based on phylogenetic classification. The receptor for HRV-C viruses is currently unknown. HRV enters host cells via endocytosis mediated by these receptors, and the process can be clathrin-dependent or independent, depending on the receptor type[14]. Once HRV enters host cells, pattern recognition receptors (PRRs) within the airway recognize HRV, which induces proinflammatory and antiviral responses. These PRRs include Toll-like receptors (TLRs), retinoic acid inducible protein I (RIG-I), and melanoma differentiation associated gene 5 (*MDA5*). In the endosome, TLR3 and TLR7/8 recognize dsRNA intermediates generated during the HRV replication cycle and ssRNA, respectively. TLR7/8 may be induced upon HRV infection, but currently TLR3 seems to be the dominant PRR for HRV. Detection of dsRNA intermediates by TLR3 triggers upregulation of cytoplasmic RIG-I and MDA5, which also recognize newly synthesized ssRNA and dsRNA intermediates in the cytoplasm[15,16] (Fig. 1). Subsequently, these 2 PRRs stimulate the interferon (IFN) gene and the expression of proinflammatory cytokines, such as RANTES, IFN-γ-induced protein 10 (IP-10), and IL-8[15,17]. These cytokines modulate proliferation, activation, and recruitment of inflammatory cells to the airway. Specifically, IL-8, a chemoattractant and activator of neutrophils, is known to be a major determinant of clinical outcomes of HRV infection. Bronchial epithelial cells (BECs) produce type I IFN-β and type III IFN-λs upon HRV infection[15,16,18]. These 2 IFN families exhibit very similar properties. Both IFN-β and IFN-λ signals through their respective receptors initiate transcription of IFN-stimulated genes (ISGs)[19,20]. The ISGs modulate HRV elimination by degrading viral RNA, preventing virus-associated protein trafficking and virion assembly, and inducing apoptosis. Limited studies suggest that a key difference between these 2 IFN families could be the cell specificity of the responses to each. Unlike the type I IFN response, which occurs in most cell types, the response to IFN-λ appears to be restricted to epithelial cells, suggesting that IFN-λ plays a dominant role in protection against viral invasion through skin and mucosal surfaces[21].

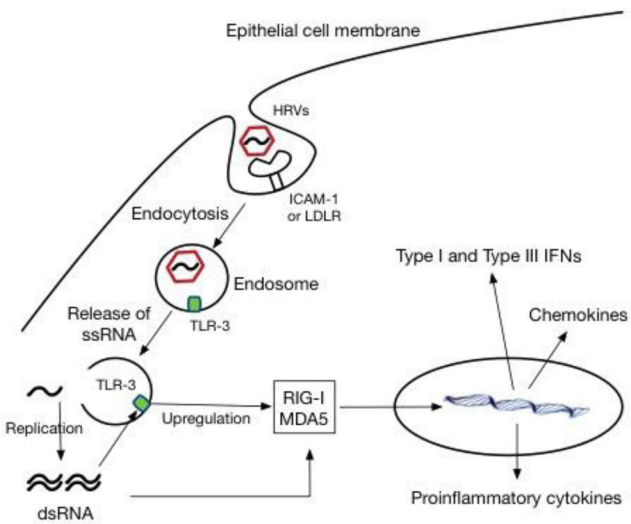

Fig. 1. Human rhinovirus (HRV) infection and innate immune responses in epithelial cells. ICAM-1, intercellular adhesion molecule 1; LDLR, low-density lipoprotein receptor; TLR-3, Toll-like receptor-3; ssRNA, single-stranded RNA; dsRNA, double-stranded RNA; RIG-I, retinoic acid inducible protein I; MDA5, melanoma differentiation associated gene 5; IFN, interferon.

Murine model of HRV infection

Until recently, the lack of small animal models of HRV infection has been a major obstacle to the investigation of the pathogenesis of HRV infection. One of the main issues confronting mouse model development was that HRVs have a high degree of species specificity. To our knowledge, there are no known murine rhinoviruses, and the major group HRVs cannot bind to ICAM-1 from other species, with the probable exception of chimpanzees and gibbons[22,23]. However, transfection of chimeric human/mouse ICAM-1 *in vitro* rendered mouse respiratory cells susceptible to HRV16, a major group HRV, infection. In contrast to major group viruses, minor group viruses can attach to mouse LDLR, but only HRV-1A and HRV-1B can replicate in mouse cells[24]. Based on the results from these *in vitro* studies, practical mouse models of major and minor group HRV infection were introduced in 2008[25]. One limitation to these murine models is that HRV titers decline rapidly within 24 hours after inoculation, preventing investigations of viral replication and airway dysfunction beyond this time point. Nonetheless, the development of rodent models of HRV infection has provided a useful tool to examine the immunopathogenesis of HRV and to understand the role of HRV in the development and acute exacerbation of

asthma *in vivo*.

Role of HRV infection in asthma development

Although asthma can arise at any age and its phenotypic expression can vary over time, asthma often originates early in life [26]. In addition to individual host factors, environmental exposures have a major impact on the development of asthma. There is little doubt that respiratory viral infections constitute an important environmental risk factor[1,3,20]. The most common viruses associated with wheezing illnesses early in life are RSV and HRV[27]. Multiple epidemiological studies have demonstrated that severe RSV bronchiolitis is associated with subsequent, persistent wheezing and childhood asthma[4-6]. With advancements in molecular assays to detect respiratory viruses, it has become apparent that HRV infections during infancy are a strong predictor for the development of asthma[28].

In this regard, the Childhood Origin of Asthma (COAST) study has provided valuable insight into the association between early life HRV infection and subsequent asthma development[29]. COAST is a high-risk birth cohort study investigating the role of immune response aberrations and respiratory viruses in the development of asthma and other allergic diseases. Data from this cohort showed that at least one wheezing illness with rhinovirus infection during infancy was the most significant risk factor for persistent wheezing at the age of 3 years[30]. Furthermore, HRV wheezing illness in the first 3 years of life was a much stronger risk factor for the development of asthma at the age of 6 years than RSV wheezing illness or aeroallergen sensitization[6]. Another high-risk birth cohort study in Australia reported similar findings: HRV wheezing illness during the first year of life was associated with increased asthma risk at the age of 5 years, although this association was restricted to children with aeroallergen sensitization by the age of 2 years[31]. However, it should be noted that these studies are based on outpatient cohorts of infants that are at high risk for developing asthma; therefore, these findings may not be applicable to the general population.

There are limited data available on the association between hospitalization with severe HRV wheezing illness and persistent wheezing or future asthma in the general population. Recent studies in Europe provide the information about this question. An Italian cohort study demonstrated that the rate of recurrent wheezing was higher after hospitalization with HRV bronchiolitis than with other respiratory viruses during a 1-year follow-up period[32]. HRV infection and a positive family history for asthma were independent risk factors for recurrent wheezing in this cohort. A case control study in Finland also demonstrated that infants hospitalized with HRV wheezing had the highest risk for subsequent asthma, and this relationship persisted at least through the adolescence[33]. Taken together, these studies suggest the importance of early life HRV infection as a major risk factor for recurrent wheezing illness and future asthma inception.

Although virtually all children (>90%) are infected with HRV during infancy[34], the factors that predispose a subset to develop persistent wheezing illnesses or asthma are not fully understood. Individual host factors, such as genetic factors and atopy, most likely interact with HRV infections in susceptible patients, resulting in an increased risk for asthma development. Evidence regarding genetic susceptibility in HRV infection is beginning to emerge. Genome-wide association studies (GWAS) have consistently linked the asthma susceptibility locus 17q21 to nonatopic childhood-onset asthma[35,36]. Data from 2 birth cohort studies, COAST and Copenhagen Studies on Asthma in Childhood, demonstrated that 17q21 variants were associated with HRV wheezing illness in early life, but not with RSV wheezing illnesses. Children with the risk-associated genetic variants only developed asthma after wheezing with HRV infection[9]. Although the disease-associated variants at this locus impact the expression levels of 2 17q21 genes, *GSDMB* and *ORMDL3*, the functions of the products encoded by these genes in the pathogenesis of asthma have not been well understood. Recently, however, potential roles of *ORMDL3* in asthma pathogenesis have been identified. These included dysregulation of the unfolded protein response, leading to airway inflammation and remodeling through sarco/endoplasmic reticulum Ca^{2+} ATPase, and negative regulation of sphingolipid synthesis resulting in airway hyperresponsiveness[37-39]. In contrast to the results from the birth cohort studies, recent *in vitro* studies have suggested that ORMDL3 might have an antiviral effect, rather than increase susceptibility to HRV infection. Overexpression of ORMDL3 in human airway epithelial cells increased the expression of 2'-5' oligoadenylate synthase[38], a molecule that inhibits viral replication by degrading viral RNA through RNase L activation[40] (Fig. 2). Moreover, the expression

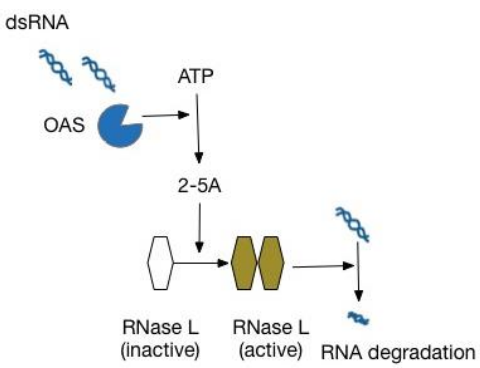

Fig. 2. The OAS/RNase L pathway in an innate immune response against viruses. dsRNA, double-stranded RNA; OAS, 2'-5'-oligoadenylate synthetase; 2-5A, 5'-phosphorylated, 2'-5'-linked oligoadenylates; RNase L, 2'-5'-oligoadenylate-dependent ribonuclease L.

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level of *ORMDL3* was significantly increased in HRV-stimulated peripheral blood mononuclear cells[9]. Therefore, the exact role of *ORMDL3* in HRV infection needs to be clarified by further epidemiological and experimental studies.

Regarding genetic susceptibility to asthma and HRV infection, a recent GWAS identified CDHR3 (encoding cadherin-related family member 3), which is highly expressed in the airway epithelium, as a new susceptibility locus for early childhood asthma [41]. A subsequent *in vitro* study suggested that mutations in this susceptibility gene could be a risk factor for increased HRV wheezing illness by mediating HRV-C entry into host cells[42]. HRV-C has been identified as the main pathogenic HRV species associated with infant wheeze, hospitalization, and subsequent development of asthma in recent research[43]. Together, this evidence suggests that HRV infection is associated with asthma susceptibility in genetically predisposed children.

Another potential host factor involved in promoting asthma inception via interaction with HRV infection is atopy. Early sensitization to allergens has consistently been identified as a major risk factor for asthma development. However, the temporal sequence of these 2 events has not been clear. Recently, however, several studies have suggested that allergic sensitization may precede HRV wheezing illness, while HRV wheezing does not increase the risk of subsequent allergic sensitization[44-46]. A number of mechanisms by which allergic sensitization might increase the risk of HRV-induced wheezing have been proposed. Th2 cytokines, such as interleukin (IL)-4, IL-5, and IL-13, further upregulate the expression of ICAM-1, the receptor for 90% of HRV serotypes, on the surface of epithelial cells in the respiratory tract[47], while HRV infection itself increases ICAM-1 expression[48]. This response may render epithelial cells more susceptible to HRV infection. Moreover, the epithelial inflammatory response to HRV is downregulated in an atopic environment, resulting in increased viral replication and cell damage, whereas transforming growth factor is upregulated[49]. These results suggest one possible mechanism by which HRV infections could promote airway remodeling. Allergic inflammation may directly inhibit host antiviral responses. An *ex vivo* model demonstrated that plasmacytoid dendritic cells (pDCs), key antigen-presenting cells recruited to the airway during viral infections, from patients with asthma secreted significantly less IFN-α upon exposure to influenza, and IgE cross-linking prior to viral challenge resulted in the abrogation of a virus-induced pDC IFN-α response[50]. This counterregulation between IgE-mediated pathways and antiviral responses might increase the severity of disease following viral infection in allergic individuals.

Supporting the "2-hit hypothesis" of asthma development, these observations have led to the notion that genetic predisposition combined with HRV infection may promote asthma in children[51]. Further research is needed to explore host factors and additional environmental factors that modify the risk for asthma following early life HRV infection and the mechanisms that underlie the interactions between HRV infection and these factors.

Impacts of HRV infection on asthma exacerbation

Respiratory viral infection has been recognized as the most common cause of asthma exacerbation[52,53]. A clear temporal relationship between outbreaks of upper respiratory viral infection and increased hospitalizations for asthma exacerbations is consistently observed in the northern hemisphere in the spring and also early fall, with a peak in September[54,55]. This seasonal pattern is sometimes referred to as the "September epidemic." Epidemic peaks of asthma exacerbations in adults are also observed shortly after children return to school and at the peak time of year for HRV infections, suggesting viral infections as a cause.

Before the advent of polymerase chain reaction (PCR) assays, the role of viral infections in asthma exacerbation was underappreciated. Molecular assays to detect respiratory viruses changed our understanding of asthma exacerbations. PCR-based studies investigating the prevalence of viral infection during asthma exacerbations showed that upper respiratory viral infections were associated with up to 85% of exacerbated asthma cases in children, and approximately 50% of these cases in adults[54,56-58]. Among viruses detected in these studies, HRV was the dominant pathogen, making up about 60% of the viruses detected[59,60]. Moreover, HRV was also the most common viral pathogen detected in children and adults hospitalized for asthma exacerbations. While the association is clear, the molecular and cellular events behind HRV-induced asthma exacerbations remain unclear.

A number of mechanisms by which HRV might exacerbate asthma have been proposed. Once, it was assumed that HRV could not infect the lower airway, because the optimal temperature for HRV replication was determined to be 33°C–35°C in early studies[61,62]. Then the common explanation was that inspiration of dry and cold air through mouth due to nasal blockage triggered asthma symptoms. Nasal-bronchial reflex and release of proinflammatory mediators from the nose into the circulation have been also proposed as underlying mechanisms.

However, it is now apparent, from subsequent *in vitro* and *in vivo* studies, that HRV can infect the lower airways. HRV has been detected in the lower airways via immunohistochemistry, reverse transcription-, and *in situ* hybridization of positive-strand viral RNA[63-65]. Interestingly, in contrast to influenza and other respiratory viruses, HRV infects relatively few epithelial cells in the airway and causes minimal cytotoxicity[66,67]. Moreover, the amount of virus shed and the extent of epithelial damage does

not correlate with the severity of symptoms[68]. Together, these studies suggest that HRV provokes symptoms through virus-specific cytopathic effects, rather than through direct cytotoxic effects.

One possible explanation for these findings is that HRV infection induces proinflammatory responses in airway epithelial cells, the primary site of HRV infection and replication. Studies using cultured human airway epithelial cells demonstrated that HRV infection induced the expression of a wide range of cytokines (IL-1β, IL-6, IL-11), growth factors (G-CSF, GM-CSF), and chemokines (CXCL8, CXCL5, CXCL10, RANTES) that can contribute to the activation and recruitment of inflammatory cells to the airway [69,70]. These cytokines were also increased in airway secretions during *in vivo* HRV infections[71]. These molecules can be expected to enhance airway inflammation and potentiate preexisting allergic inflammation.

In addition to the pro-inflammatory cytokines mentioned above, HRV infection can trigger the release of Th2-promoting cytokines, such as IL-25, IL-33, and thymic stromal lymphopoietin (TSLP), from airway epithelial cells. These cytokines have been recognized as critical elements in type 2 T-cell differentiation. Recently, a number of studies have provided information about the role of these epithelium-derived cytokines in HRV-induced asthma exacerbation. Human *in vitro* and *in vivo* HRV infection studies have demonstrated a heightened intrinsic capacity for IL-25 expression in HRV-infected asthmatic BECs and increased IL-25 level in the nasal fluid of atopic asthmatics in experimental HRV infections. Similar findings were observed in a murine model of HRV-induced asthma exacerbations. In addition, blocking the IL-25 receptor reduced type 2 cytokine expression, mucus production, and recruitment of inflammatory cells[72]. Another human experimental HRV infection model revealed increased levels of IL-33 and type 2 cytokines in the bronchial mucosal lining fluid of asthmatics during experimental HRV infection, and the levels of these cytokines were related to the severity of exacerbation. The production of these cytokines by peripheral blood T cells and type 2 innate lymphoid cells from the same subjects was induced when cultured with the supernatants of HRV-infected BECs and this induction was entirely dependent upon IL-33[73]. TSLP is also considered a potential molecular link between HRV infection and the characteristic Th2 response seen in asthma exacerbation. An *ex vivo* model using asthmatic BECs revealed that stimulation with dsRNA, a surrogate of viral infection, induced exaggerated TSLP production, accompanied by reduced IFN-β production[74]. Furthermore, lung tissue TSLP was induced by both HRV infection and exposure to dsRNA in a murine model of allergic asthma[75]. Taken together, these observations suggest a potential role of epithelial mediators in HRV-induced asthma exacerbation.

IFNs play a critical role in the host defense against respiratory viruses, including HRV. Type I (IFN-α/β) and type III (IFN-λ) INFs are the most significant IFNs secreted by the airway epithelium during acute HRV infection[21,76]. It has been suggested that impaired epithelial production of IFN-β and IFN-λ in asthmatic subjects may contribute to viral exacerbation of asthma[77,78]. Recent studies investigating airway interferon production in response to HRV in children have demonstrated reduced IFN secretion from BECs after HRV stimulation in asthmatic and atopic nonasthmatic children compared with IFN secretion seen in nonatopic, nonasthmatic children[79]. Additionally, *ex vivo* IFN-λ and IFN-β induction correlated inversely with severity of symptoms, bronchoalveolar lavage viral load, and airway inflammation, and correlated positively with reductions in lung function during *in vivo* HRV infection[78]. However, the association between IFN response and HRV infection in asthmatic children seems to be more complex. A prospective study in school-aged children with asthma showed increased IFN-λ1 production in wheezing children with HRV infection as compared to nonwheezing children with HRV infection, suggesting that a dysregulated IFN response rather than a simple deficiency of IFN production mediates HRV-induced asthma exacerbations[80]. Further research is needed to define role of IFN-λ in HRV-induced asthma exacerbation and the mechanism of this IFN dysregulation.

HRV infections might also contribute to asthma exacerbations via enhanced mucus production. *In vitro* studies have shown that HRV infection of human airway epithelial cells increased mRNA expression of mucin-2 (Muc2), mucin-3 (Muc3), mucin-5 subtypes A and C (Muc5AC), mucin-5 subtype B (Muc5B), and mucin-6 (Muc6). MUC5AC and total mucin concentrations increased in supernatants and lysates of the surface cells[81]. Supporting this *in vitro* study, a murine model of HRV infection demonstrated that RV1B induced Muc5AC and Muc5B mRNA in the lung, and increased MUC5AC and MUC5B protein in BAL[25].

HRV infections can affect the contractility of airway smooth muscle cells, which might contribute to asthma exacerbations. Limited *in vitro* and animal studies have revealed that binding of HRV to its receptor, ICAM-1, on airway smooth muscle cell surfaces can enhance the contractility of airway smooth muscle cells and impair the response to β-adrenergic agonists without any cytopathic effects or viral replication[82,83]. Moreover, several *in vitro* studies have demonstrated that HRV can infect a variety of other cell types, including fibroblasts, monocytes, and macrophages, although the contributions of these individual cell types to asthma exacerbation are still not well understood. Further research is needed to define the role of these cells in HRV-induced asthma exacerbation.

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Conclusion

The advent of molecular diagnostics to detect respiratory viruses has led to new insights into the role of HRV infections in the development and acute exacerbation of asthma in children. Recent clinical and experimental studies have identified plausible pathways by which HRV infection could be causal, particularly in a susceptible host and trigger disease exacerbation. Airway epithelial cells, the primary site of infection and replication of HRV, play a key role in these processes. Additional *in vitro* and *in vivo* studies using murine model of HRV infection with genetically modified mouse may help to improve our understanding of the interplay between genetic factors and HRV infection in the development of childhood asthma and to identify novel targets for therapeutic intervention.

Conflict of interest

No potential conflict of interest relevant to this article was reported.

References

1. Duijts L. Fetal and infant origins of asthma. Eur J Epidemiol 2012;27:5-14.
2. Le Souëf PN. Gene-environmental interaction in the development of atopic asthma: new developments. Curr Opin Allergy Clin Immunol 2009;9:123-7.
3. von Mutius E. Environmental factors influencing the development and progression of pediatric asthma. J Allergy Clin Immunol 2002;109(6 Suppl):S525-32.
4. Stein RT, Sherrill D, Morgan WJ, Holberg CJ, Halonen M, Taussig LM, et al. Respiratory syncytial virus in early life and risk of wheeze and allergy by age 13 years. Lancet 1999;354:541-5.
5. Henderson J, Hilliard TN, Sherriff A, Stalker D, Al Shammari N, Thomas HM. Hospitalization for RSV bronchiolitis before 12 months of age and subsequent asthma, atopy and wheeze: a longitudinal birth cohort study. Pediatr Allergy Immunol 2005;16:386-92.
6. Jackson DJ, Gangnon RE, Evans MD, Roberg KA, Anderson EL, Pappas TE, et al. Wheezing rhinovirus illnesses in early life predict asthma development in high-risk children. Am J Respir Crit Care Med 2008;178:667-72.
7. Gern JE, Busse WW. Relationship of viral infections to wheezing illnesses and asthma. Nat Rev Immunol 2002;2:132-8.
8. Koponen P, Helminen M, Paassilta M, Luukkaala T, Korppi M. Preschool asthma after bronchiolitis in infancy. Eur Respir J 2012;39:76-80.
9. Calıskan M, Bochkov YA, Kreiner-Møller E, Bønnelykke K, Stein MM, Du G, et al. Rhinovirus wheezing illness and genetic risk of childhood-onset asthma. N Engl J Med 2013;368:1398-407.
10. Palmenberg AC, Spiro D, Kuzmickas R, Wang S, Djikeng A, Rathe JA, et al. Sequencing and analyses of all known human rhinovirus genomes reveal structure and evolution. Science 2009;324:55-9.
11. Bochkov YA, Palmenberg AC, Lee WM, Rathe JA, Amineva SP, Sun X, et al. Molecular modeling, organ culture and reverse genetics for a newly identified human rhinovirus C. Nat Med 2011;17:627-32.
12. Greve JM, Davis G, Meyer AM, Forte CP, Yost SC, Marlor CW, et al. The major human rhinovirus receptor is ICAM-1. Cell 1989;56:839-47.
13. Staunton DE, Merluzzi VJ, Rothlein R, Barton R, Marlin SD, Springer TA. A cell adhesion molecule, ICAM-1, is the major surface receptor for rhinoviruses. Cell 1989;56:849-53.
14. Dreschers S, Dumitru CA, Adams C, Gulbins E. The cold case: are rhinoviruses perfectly adapted pathogens? Cell Mol Life Sci 2007;64:181-91.
15. Slater L, Bartlett NW, Haas JJ, Zhu J, Message SD, Walton RP, et al. Co-ordinated role of TLR3, RIG-I and MDA5 in the innate response to rhinovirus in bronchial epithelium. PLoS Pathog 2010;6:e1001178.
16. Wang Q, Nagarkar DR, Bowman ER, Schneider D, Gosangi B, Lei J, et al. Role of double-stranded RNA pattern recognition receptors in rhinovirus-induced airway epithelial cell responses. J Immunol 2009;183:6989-97.
17. Triantafilou K, Vakakis E, Richer EA, Evans GL, Villiers JP, Triantafilou M. Human rhinovirus recognition in non-immune cells is mediated by Toll-like receptors and MDA-5, which trigger a synergetic pro-inflammatory immune response. Virulence 2011;2:22-9.
18. Khaitov MR. Acute respiratory viral infections and bronchial asthma. Cellular and molecular aspects of the problem. Zh Mikrobiol Epidemiol Immunobiol 2002;(4):84-93.
19. Bochkov YA, Hanson KM, Keles S, Brockman-Schneider RA, Jarjour NN, Gern JE. Rhinovirus-induced modulation of gene expression in bronchial epithelial cells from subjects with asthma. Mucosal Immunol 2010;3:69-80.
20. Proud D, Turner RB, Winther B, Wiehler S, Tiesman JP, Reichling TD, et al. Gene expression profiles during in vivo human rhinovirus infection: insights into the host response. Am J Respir Crit Care Med 2008;178:962-8.
21. Sommereyns C, Paul S, Staeheli P, Michiels T. IFN-lambda (IFN-lambda) is expressed in a tissue-dependent fashion and primarily acts on epithelial cells in vivo. PLoS Pathog 2008;4:e1000017.
22. Dick EC. Experimental infections of chimpanzees with human rhinovirus types 14 and 43. Proc Soc Exp Biol Med 1968;127:1079-81.
23. Pinto CA, Haff RF. Experimental infection of gibbons with rhinovirus. Nature 1969;224:1310-1.
24. Tuthill TJ, Papadopoulos NG, Jourdan P, Challinor LJ, Sharp NA, Plumpton C, et al. Mouse respiratory epithelial cells support efficient replication of human rhinovirus. J Gen Virol 2003;84(Pt 10):2829-36.
25. Bartlett NW, Walton RP, Edwards MR, Aniscenko J, Caramori G, Zhu J, et al. Mouse models of rhinovirus-induced disease and exacerbation of allergic airway inflammation. Nat Med 2008;14:199-204.
26. Carraro S, Scheltema N, Bont L, Baraldi E. Early-life origins of chronic respiratory diseases: understanding and promoting healthy ageing. Eur Respir J 2014;44:1682-96.
27. Jackson DJ, Lemanske RF Jr. The role of respiratory virus infections in childhood asthma inception. Immunol Allergy Clin North Am 2010;30:513-22.
28. Kotaniemi-Syrjänen A, Vainionpää R, Reijonen TM, Waris M, Korhonen K, Korppi M. Rhinovirus-induced wheezing in infancy: the first sign of childhood asthma? J Allergy Clin Immunol 2003;

111:66-71.
29. Lemanske RF Jr. The childhood origins of asthma (COAST) study. Pediatr Allergy Immunol 2002;13 Suppl 15:38-43.
30. Lemanske RF Jr, Jackson DJ, Gangnon RE, Evans MD, Li Z, Shult PA, et al. Rhinovirus illnesses during infancy predict subsequent childhood wheezing. J Allergy Clin Immunol 2005;116:571-7.
31. Kusel MM, de Klerk NH, Kebadze T, Vohma V, Holt PG, Johnston SL, et al. Early-life respiratory viral infections, atopic sensitization, and risk of subsequent development of persistent asthma. J Allergy Clin Immunol 2007;119:1105-10.
32. Midulla F, Pierangeli A, Cangiano G, Bonci E, Salvadei S, Scagnolari C, et al. Rhinovirus bronchiolitis and recurrent wheezing: 1-year follow-up. Eur Respir J 2012;39:396-402.
33. Hyvärinen MK, Kotaniemi-Syrjänen A, Reijonen TM, Korhonen K, Korppi MO. Teenage asthma after severe early childhood wheezing: an 11-year prospective follow-up. Pediatr Pulmonol 2005; 40:316-23.
34. Kieninger E, Fuchs O, Latzin P, Frey U, Regamey N. Rhinovirus infections in infancy and early childhood. Eur Respir J 2013;41: 443-52.
35. Akhabir L, Sandford AJ. Genome-wide association studies for discovery of genes involved in asthma. Respirology 2011;16:396-406.
36. Kang MJ, Yu HS, Seo JH, Kim HY, Jung YH, Kim YJ, et al. GSDMB/ORMDL3 variants contribute to asthma susceptibility and eosinophil-mediated bronchial hyperresponsiveness. Hum Immunol 2012;73:954-9.
37. Mahn K, Hirst SJ, Ying S, Holt MR, Lavender P, Ojo OO, et al. Diminished sarco/endoplasmic reticulum Ca2+ ATPase (SERCA) expression contributes to airway remodelling in bronchial asthma. Proc Natl Acad Sci U S A 2009;106:10775-80.
38. Miller M, Tam AB, Cho JY, Doherty TA, Pham A, Khorram N, et al. ORMDL3 is an inducible lung epithelial gene regulating metalloproteases, chemokines, OAS, and ATF6. Proc Natl Acad Sci U S A 2012;109:16648-53.
39. Worgall TS, Veerappan A, Sung B, Kim BI, Weiner E, Bholah R, et al. Impaired sphingolipid synthesis in the respiratory tract induces airway hyperreactivity. Sci Transl Med 2013;5:186ra67.
40. Choi UY, Kang JS, Hwang YS, Kim YJ. Oligoadenylate synthase-like (OASL) proteins: dual functions and associations with diseases. Exp Mol Med 2015;47:e144.
41. Bonnelykke K, Sleiman P, Nielsen K, Kreiner-Møller E, Mercader JM, Belgrave D, et al. A genome-wide association study identifies CDHR3 as a susceptibility locus for early childhood asthma with severe exacerbations. Nat Genet 2014;46:51-5.
42. Bochkov YA, Watters K, Ashraf S, Griggs TF, Devries MK, Jackson DJ, et al. Cadherin-related family member 3, a childhood asthma susceptibility gene product, mediates rhinovirus C binding and replication. Proc Natl Acad Sci U S A 2015;112:5485-90.
43. Cox DW, Bizzintino J, Ferrari G, Khoo SK, Zhang G, Whelan S, et al. Human rhinovirus species C infection in young children with acute wheeze is associated with increased acute respiratory hospital admissions. Am J Respir Crit Care Med 2013;188:1358-64.
44. Jackson DJ, Evans MD, Gangnon RE, Tisler CJ, Pappas TE, Lee WM, et al. Evidence for a causal relationship between allergic sensitization and rhinovirus wheezing in early life. Am J Respir Crit Care Med 2012;185:281-5.
45. Jackson DJ. The role of rhinovirus infections in the development of early childhood asthma. Curr Opin Allergy Clin Immunol 2010; 10:133-8.
46. Jartti T, Kuusipalo H, Vuorinen T, Söderlund-Venermo M, Allander T, Waris M, et al. Allergic sensitization is associated with rhinovirus-, but not other virus-, induced wheezing in children. Pediatr Allergy Immunol 2010;21:1008-14.
47. Bianco A, Sethi SK, Allen JT, Knight RA, Spiteri MA. Th2 cytokines exert a dominant influence on epithelial cell expression of the major group human rhinovirus receptor, ICAM-1. Eur Respir J 1998;12:619-26.
48. Winther B, Arruda E, Witek TJ, Marlin SD, Tsianco MM, Innes DJ, et al. Expression of ICAM-1 in nasal epithelium and levels of soluble ICAM-1 in nasal lavage fluid during human experimental rhinovirus infection. Arch Otolaryngol Head Neck Surg 2002;128: 131-6.
49. Xatzipsalti M, Psarros F, Konstantinou G, Gaga M, Gourgiotis D, Saxoni-Papageorgiou P, et al. Modulation of the epithelial inflammatory response to rhinovirus in an atopic environment. Clin Exp Allergy 2008;38:466-72.
50. Gill MA, Bajwa G, George TA, Dong CC, Dougherty II, Jiang N, et al. Counterregulation between the FcepsilonRI pathway and antiviral responses in human plasmacytoid dendritic cells. J Immunol 2010;184:5999-6006.
51. Gern JE. The ABCs of rhinoviruses, wheezing, and asthma. J Virol 2010;84:7418-26.
52. Busse WW, Lemanske RF Jr, Gern JE. Role of viral respiratory infections in asthma and asthma exacerbations. Lancet 2010;376: 826-34.
53. Johnston SL, Pattemore PK, Sanderson G, Smith S, Lampe F, Josephs L, et al. Community study of role of viral infections in exacerbations of asthma in 9-11 year old children. BMJ 1995;310: 1225-9.
54. Heymann PW, Carper HT, Murphy DD, Platts-Mills TA, Patrie J, McLaughlin AP, et al. Viral infections in relation to age, atopy, and season of admission among children hospitalized for wheezing. J Allergy Clin Immunol 2004;114:239-47.
55. Johnston NW, Johnston SL, Norman GR, Dai J, Sears MR. The September epidemic of asthma hospitalization: school children as disease vectors. J Allergy Clin Immunol 2006;117:557-62.
56. Mallia P, Johnston SL. How viral infections cause exacerbation of airway diseases. Chest 2006;130:1203-10.
57. Kling S, Donninger H, Williams Z, Vermeulen J, Weinberg E, Latiff K, et al. Persistence of rhinovirus RNA after asthma exacerbation in children. Clin Exp Allergy 2005;35:672-8.
58. Proud D, Chow CW. Role of viral infections in asthma and chronic obstructive pulmonary disease. Am J Respir Cell Mol Biol 2006;35: 513-8.
59. Jackson DJ, Johnston SL. The role of viruses in acute exacerbations of asthma. J Allergy Clin Immunol 2010;125:1178-87.
60. Khetsuriani N, Kazerouni NN, Erdman DD, Lu X, Redd SC, Anderson LJ, et al. Prevalence of viral respiratory tract infections in children with asthma. J Allergy Clin Immunol 2007;119:314-21.
61. Stott EJ, Heath GF. Factors affecting the growth of Rhinovirus 2 in suspension cultures of L132 cells. J Gen Virol 1970;6:15-24.
62. Papadopoulos NG, Sanderson G, Hunter J, Johnston SL. Rhinoviruses replicate effectively at lower airway temperatures. J Med Virol 1999;58:100-4.
63. Papadopoulos NG, Bates PJ, Bardin PG, Papi A, Leir SH, Fraenkel DJ, et al. Rhinoviruses infect the lower airways. J Infect Dis 2000; 181:1875-84.
64. Gern JE, Galagan DM, Jarjour NN, Dick EC, Busse WW. Detection of rhinovirus RNA in lower airway cells during experimentally induced infection. Am J Respir Crit Care Med 1997;155:1159-61.
65. Wos M, Sanak M, Soja J, Olechnowicz H, Busse WW, Szczeklik A. The presence of rhinovirus in lower airways of patients with

bronchial asthma. Am J Respir Crit Care Med 2008;177:1082-9.
66. Arruda E, Boyle TR, Winther B, Pevear DC, Gwaltney JM Jr, Hayden FG. Localization of human rhinovirus replication in the upper respiratory tract by in situ hybridization. J Infect Dis 1995;171:1329-33.
67. Winther B, Brofeldt S, Christensen B, Mygind N. Light and scanning electron microscopy of nasal biopsy material from patients with naturally acquired common colds. Acta Otolaryngol 1984;97:309-18.
68. Turner RB, Hendley JO, Gwaltney JM Jr. Shedding of infected ciliated epithelial cells in rhinovirus colds. J Infect Dis 1982;145:849-53.
69. Chen Y, Hamati E, Lee PK, Lee WM, Wachi S, Schnurr D, et al. Rhinovirus induces airway epithelial gene expression through double-stranded RNA and IFN-dependent pathways. Am J Respir Cell Mol Biol 2006;34:192-203.
70. Spurrell JC, Wiehler S, Zaheer RS, Sanders SP, Proud D. Human airway epithelial cells produce IP-10 (CXCL10) in vitro and in vivo upon rhinovirus infection. Am J Physiol Lung Cell Mol Physiol 2005;289:L85-95.
71. Proud D, Gwaltney JM Jr, Hendley JO, Dinarello CA, Gillis S, Schleimer RP. Increased levels of interleukin-1 are detected in nasal secretions of volunteers during experimental rhinovirus colds. J Infect Dis 1994;169:1007-13.
72. Beale J, Jayaraman A, Jackson DJ, Macintyre JD, Edwards MR, Walton RP, et al. Rhinovirus-induced IL-25 in asthma exacerbation drives type 2 immunity and allergic pulmonary inflammation. Sci Transl Med 2014;6:256ra134.
73. Jackson DJ, Makrinioti H, Rana BM, Shamji BW, Trujillo-Torralbo MB, Footitt J, et al. IL-33-dependent type 2 inflammation during rhinovirus-induced asthma exacerbations in vivo. Am J Respir Crit Care Med 2014;190:1373-82.
74. Uller L, Leino M, Bedke N, Sammut D, Green B, Lau L, et al. Double-stranded RNA induces disproportionate expression of thymic stromal lymphopoietin versus interferon-beta in bronchial epithelial cells from donors with asthma. Thorax 2010;65:626-32.
75. Mahmutovic-Persson I, Akbarshahi H, Bartlett NW, Glanville N, Johnston SL, Brandelius A, et al. Inhaled dsRNA and rhinovirus evoke neutrophilic exacerbation and lung expression of thymic stromal lymphopoietin in allergic mice with established experimental asthma. Allergy 2014;69:348-58.
76. Lewis TC, Henderson TA, Carpenter AR, Ramirez IA, McHenry CL, Goldsmith AM, et al. Nasal cytokine responses to natural colds in asthmatic children. Clin Exp Allergy 2012;42:1734-44.
77. Wark PA, Johnston SL, Bucchieri F, Powell R, Puddicombe S, Laza-Stanca V, et al. Asthmatic bronchial epithelial cells have a deficient innate immune response to infection with rhinovirus. J Exp Med 2005;201:937-47.
78. Contoli M, Message SD, Laza-Stanca V, Edwards MR, Wark PA, Bartlett NW, et al. Role of deficient type III interferon-lambda production in asthma exacerbations. Nat Med 2006;12:1023-6.
79. Baraldo S, Contoli M, Bazzan E, Turato G, Padovani A, Marku B, et al. Deficient antiviral immune responses in childhood: distinct roles of atopy and asthma. J Allergy Clin Immunol 2012;130:1307-14.
80. Miller EK, Hernandez JZ, Wimmenauer V, Shepherd BE, Hijano D, Libster R, et al. A mechanistic role for type III IFN-λ1 in asthma exacerbations mediated by human rhinoviruses. Am J Respir Crit Care Med 2012;185:508-16.
81. Inoue D, Yamaya M, Kubo H, Sasaki T, Hosoda M, Numasaki M, et al. Mechanisms of mucin production by rhinovirus infection in cultured human airway epithelial cells. Respir Physiol Neurobiol 2006;154:484-99.
82. Nagarkar DR, Bowman ER, Schneider D, Wang Q, Shim J, Zhao Y, et al. Rhinovirus infection of allergen-sensitized and -challenged mice induces eotaxin release from functionally polarized macrophages. J Immunol 2010;185:2525-35.
83. Grunstein MM, Hakonarson H, Whelan R, Yu Z, Grunstein JS, Chuang S. Rhinovirus elicits proasthmatic changes in airway responsiveness independently of viral infection. J Allergy Clin Immunol 2001;108:997-1004.