Do early-life viral infections cause asthma?

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Epidemiologic associations between viral lower respiratory infections (LRIs) and asthma in later childhood are well known. However, the question of whether such infections cause asthma or unmask asthma in a susceptible host has still not been settled. Most early evidence centered on the role of the respiratory syncytial virus; however, recent studies highlight a potential role for human rhinovirus as a risk factor for asthma. The links between early-life viral LRI and subsequent asthma are generally via wheeze; however, the presence of wheeze does not give any information about why the child is wheezing. Wheeze in early life is, at best, a fuzzy phenotype and not specific for subsequent asthma. The risk of asthma after viral LRI is increased in the presence of allergic sensitization in early life and if the infection is more severe. Atopy-associated mechanisms also appear to be involved in viral-induced acute exacerbations of asthma, especially in prolonging symptomatology after the virus has been cleared from the lungs. Breaking the nexus between viral respiratory infections and asthma may be possible with interventions designed to inhibit atopy-related effectors mechanisms from participating in the host response to respiratory viral infections. (J Allergy Clin Immunol 2010;125:1202-5.)

Key words: *Allergic sensitization, human rhinovirus, IgE, respiratory syncytial virus, T cells, wheeze*

Do viral respiratory infections cause asthma? The answer to this question has troubled pediatricians, respiratory researchers, and epidemiologists for many years. Much of the debate has centered on whether viral infections in early life cause asthma by damaging the developing respiratory and immune systems or whether these infections simply unmask the susceptibility for asthma in genetically predisposed infants. In addition, debate continues whether specific viruses are involved or whether the reported associations simply reflect the epidemiology of respiratory viral infections in early life. These questions have still not been resolved. This review attempts to summarize the evidence and point the way forward.

Abbreviations used
BEC: Bronchial epithelial cell
CAS: Childhood Asthma Study
HRV: Human rhinovirus
LRI: Lower respiratory infection
OR: Odds ratio
RSV: Respiratory syncytial virus
wLRI: Wheezy lower respiratory infection

WHAT HAVE WE LEARNED FROM THE EPIDEMIOLOGISTS?

The link between viral respiratory infections in early life and subsequent childhood asthma has been the subject of much study and debate for decades. Although much of the earlier literature centered on the link between hospitalization for acute viral bronchiolitis, most commonly because of infection with the respiratory syncytial virus (RSV), recent investigations into wheezing in childhood have included more comprehensive viral assessments using modern molecular techniques in community-based cohorts, including longitudinal birth cohorts. The overall conclusion from the vast majority of earlier studies was that hospitalization for RSV bronchiolitis was associated with recurrent wheeze throughout childhood but not with an increased prevalence of atopy. The 1 major exception to this pattern was the cohort of hospitalized children from Sweden studied by Sigurs et al[1] in which an increased risk for allergic sensitization at age 7 years was also reported. Wheeze after RSV bronchiolitis generally abates by the early teen years. The first of the longitudinal community-based studies adequately to address prospectively the link between respiratory viral infections in early life and subsequent asthma was the Tucson Children's Respiratory Study. Major outcomes from this study have included demonstrating the link between low lung function in early life and the risk of subsequent childhood wheeze; showing that the majority of infants who wheeze will not have asthma in later childhood; showing that RSV lower respiratory infections (LRIs) before 3 years of age are no longer a risk factor for current wheeze at age 11 to 13 years; showing that RSV LRIs before 3 years of age are associated with a lower FEV_1 at age 13 years that normalizes after bronchodilator inhalation, consistent with an abnormality in regulation of airway smooth muscle tone; and showing that chronic asthma persisting into adulthood is associated with persistent wheezing in early life, asthma at age 6 years, sensitization to *Alternaria* sp (the predominant local aeroallergen), low lung function at 6 years, and bronchial hyperresponsiveness at age 6 years.[2]

Systematic studies in animal models, supported by more limited data from human infants (see review[3]), have suggested that the age of first infection with RSV has an impact on the immunologic response to the initial and subsequent infections. In general, these studies demonstrated higher levels of T_H2 cytokines

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(IL-13 in mice and IL-4 in infants) both during the initial infection in early life and on subsequent reinfection (in mice). These data are consistent with the response of a less mature immune system; however, whether the immune response is altered by the viral infection or whether this pattern is related to the developmental stage of the immune system at the time of infection is not clear.

Since the framing of the hygiene hypothesis, which initially related the increase in the prevalence of asthma and atopy to a reduction in the prevalence of infections, much confusion has been propagated in the literature. One interpretation of this hypothesis raised the notion that viral respiratory infection may protect against the development of asthma. Authors have variably used daycare attendance and family size as surrogates for exposure to viral infections and have reported contradictory data. Few epidemiology studies have verified the respiratory infections or distinguished between infections of the upper and lower airways. We reported data from a longitudinal birth cohort study undertaken in Perth, Australia (the Western Australian Pregnancy Cohort), demonstrating that up to 3 upper respiratory infections in the first year of life were associated with a lower risk of current asthma at 6 years of age. In this cohort, LRI associated with wheeze (wLRIs) in the first year of life increased the risk of asthma in a dose-dependant fashion. These data suggest that much of the earlier epidemiology was misinterpreted; wLRIs in early life are associated with increased risk for subsequent asthma, with the evidence stronger when more precise methods are used for verifying respiratory illnesses (see review[4]).

Although the earlier studies concentrated on the links between RSV infections, either verified or assumed from the clinical pattern, and subsequent asthma, recent data have shown increased risk of asthma with LRIs caused by other viruses, especially human rhinovirus (HRV). Traditionally, HRV infections were assumed to be limited to the upper airways, where the lower temperature favored replication. However, there is now strong evidence (see review[5]) that HRV spreads to the lower airways; can infect and replicate in lower airway cells, including respiratory epithelial cells and fibroblasts; and induces cytokines responses in the lungs that may add to symptomatology. Data from 2 birth cohort studies, both undertaken in infants at high risk of developing allergies and asthma, have demonstrated strong associations between HRV-associated wLRIs and subsequent asthma. The Childhood Origins of ASThma (COAST) cohort, Madison, Wis, consists of 259 children followed prospectively from birth. In this study, wLRI with RSV (odds ratio [OR], 2.6), HRV (OR, 9.8), or both (OR, 10) was associated with asthma at 6 years of age.[6] The Childhood Asthma Study (CAS) undertaken in Perth, Australia, has reported data from 198 children followed from birth[7] showing that wheezing associated with HRV or RSV in the first year of life was a significant risk factor (OR, 2.5) for current wheeze at 5 years of age. However, the CAS study also highlighted the important role played by allergic sensitization in that the association between virus-associated wLRI and subsequent asthma was restricted to those children who had developed allergic sensitization by 2 years of age, with the OR approximately doubling in this subgroup.[7] With continued follow-up of this cohort, the association between viral-associated wLRI and current asthma, restricted to those with early allergic sensitization, holds true out to 10 years of age (unpublished observations, Kusel, December 2009).

Data from a number of longitudinal studies (see review[4]) have demonstrated that the interaction between early-life viral infections and allergic sensitization increases the risk of subsequent asthma. Most studies demonstrate independent effects for allergic sensitization and early-life wLRI on the risk for subsequent asthma and support at least an additive effect for both risk factors. In the community-based Western Australian Pregnancy Cohort, the risk of current asthma at 6 years of age was approximately doubled by the presence of sensitization to common aeroallergens at age 6 years. The risk for asthma associated with ≥2 wLRIs in the first year of life was increased 4-fold; however, the risk increased to approximately 9-fold for children who had both atopy at 6 years and ≥2 wLRIs in the first year of life. Similar data are available from cohort studies from other countries. Another factor that has emerged from these studies is that the degree of atopy—that is, atopy as a quantitative variable—appears to play an important part in the increased the risk of subsequent asthma associated with early-life wLRI (see review[4]).

SUSCEPTIBLE HOST, VIRUS-INDUCED DAMAGE, OR BOTH?

Cogent arguments can, and have been, mounted for the association between early-life viral respiratory infections and childhood asthma being caused by the virus unmasking a susceptible host, or the virus causing damage to the airways that induces remodeling and results in asthma (see review[5]). In physiological terms, the presence of wheeze simply implies the presence of flow limitation during expiration. Wheeze can result from small airways (eg, resulting from adverse *in utero* exposures that limit airway growth such as maternal smoking and household chemical exposure), narrow airways (eg, airways narrowed by mucosal edema or bronchospasm), or floppy airways (eg, airways with increased wall compliance as may be seen with premature birth or bronchomalacia). Low premorbid lung function increases the risk of wheezing with viral LRI but does not necessarily increase the risk of asthma in later childhood or adulthood.[2] In addition, a decreased ability of the infant immune system to mount an adequate antiviral defence, either through a prolonged T_H2-biased response, a decreased innate immune response, or defects in other aspects of local immunity (see reviews[4,5]), increases the likelihood of virus spreading to the lower airways and results in more severe LRI. Delayed maturation of various aspects of T_H1 function increases the risk of allergic sensitization and of severe LRI (see review[8]). Together these can result in airway inflammation at a crucial stage in lung development, which can, theoretically, initiate the process of airway remodeling. We have proposed that delayed immune maturation acts as a common predisposition for early-life wLRI, for early allergic sensitization, and for the increased risk of childhood asthma (see review[8]). In the CAS study, we measured the ability of cord blood mononuclear cells to produce T_H1 and T_H2 cytokines to mitogen stimulation. Children with a high IL-5 to IL-10 ratio had a greater risk of LRI over the first 5 years of life.[9] Diminished IL-10–mediated immune regulation is also thought to occur in patients with asthma in response to HRV LRI (see review[10]). However, additional gene expression profiling studies suggest that the $IL\text{-}5^{hi}/IL\text{-}10^{lo}$ phenotype characteristic of the at-risk infants may be a surrogate for a more complex underlying immunophenotype that included a deficiency in capacity to produce IL-21,[9] which has been identified as central to resistance to persistent viral infection.[11]

The common genetic risks for both LRI and asthma have recently been reviewed.[10] Genetic variations in genes encoding

Toll-like receptors, CD14, and other components of the host antiviral response have also been associated with various asthma phenotypes, supporting the notion of a common genetic predisposition for LRI and asthma.

Epigenetic phenomena, in which gene expression is altered without altering genetic structure, could also contribute to a common susceptibility to LRI and asthma. Evidence is emerging (see review[12]) of epigenetic regulation of airway inflammation at various steps along the inflammatory cascade. Expression of key genes in the T-cell system associated with regulation of the atopic asthmatic phenotype, exemplified by IFN-γ, are tightly controlled by promoter methylation.[13] We have presented some evidence suggesting that methylation patterns may vary between atopic and nonatopic children.[14] Epigenetic regulation can occur via a variety of mechanisms, including DNA methylation; histone modifications, either though acetylation or methylation; or modification of nuclear chaperone proteins. Tobacco smoking is thought to induce a state of relative steroid resistance in adult patients with asthma via epigenetic mechanisms. Maternal smoking during pregnancy is known to have a number of adverse effects that increase the risk of both infections in early life and childhood asthma, including altered lung growth, delayed immune maturation at birth and in the early postnatal period, and reduced postnatal vaccine responses. In addition, smoking during pregnancy increases the risk of asthma in the children of female offspring of maternal smokers, a third-generational effect. However, just how many of these adverse effects of maternal smoking during pregnancy are a result of epigenetic phenomena is not known.

In a recent departure from most literature, Thomsen et al[15] suggested that the direction of the association between RSV bronchiolitis and asthma was opposite to that previous published—that is, that asthma caused bronchiolitis. They reported data from all twins born in Denmark between 1994 and 2000, linking hospitalization as a result of RSV in early life, obtained from national records, to reported asthma from either hospital discharge records or parent-completed questionnaires when the children were age 3 to 9 years. Complete data were obtained from 5154 twin pairs. A total of 1019 children were recorded as having been hospitalized for an RSV-associated illness, with 50% admitted in the first 6 months of life. It was not possible from their report to determine how many children were classified as having asthma. In addition, neither the clinical characterization of RSV infection (upper vs lower airway; mild vs severe) nor of asthma (age range, 3-9 years; nonvalidated parent filled out questionnaire asking a single question on "asthma ever") is sufficient to give certainty to the authors' interpretation of their data. At best these data support the notion of a shared susceptibility, as discussed.

DOES THE RESPONSE TO A RESPIRATORY VIRAL INFECTION DIFFER IN THOSE WITH ASTHMA?

When respiratory viruses enter the lower airway, they infect bronchial epithelial cells (BECs) triggering an antiviral response aimed at containing and eliminating the infection. Infected BECs secrete a variety of products, including IFNs and proinflammatory cytokines such as IL-8. Recent reports demonstrated that the type III IFN, IFN-λ, is the major interferon secreted by infected BECs, followed by the type I IFN, IFN-β.[16] These IFNs induce the BEC to undergo apoptosis, thus limiting viral spread. Atopic patients with asthma have been shown to be more susceptible to infection with respiratory infections, especially with HRV, and have also been reported to have deficient production of IFN-β[17] and IFN-λ.[18] The deficiency of IFN-β results in deficient apoptosis and increased viral release from BECs cultured *in vitro*.[17] Although these data add to the body of data supporting a common susceptibility to respiratory viral infections and asthma, confirmatory data from infants and young children are lacking. Martinez[19] has recently pointed out that the data from the Tucson Children's Respiratory Study are consistent with an abnormality in the innate immune responses to viral LRI in early life. Unfortunately, that study was conducted without the benefit of modern molecular virology methods. Further longitudinal studies using such methods, together with sophisticated immunology, will be required to confirm the mechanisms underlying a common susceptibility to respiratory viral infections and asthma.

Many studies have demonstrated the role of respiratory viruses, especially HRV, in inducing acute exacerbations of asthma (see review[3]). Recent data from our group[20] have reported a mechanism that could explain the prolonged symptomatology associated with virus-induced acute exacerbations of asthma, airway inflammation, and airway hyperresponsiveness, which can persist long after viral clearance. Collected by using a genomics-based approach profiling PBMCs collected during the acute exacerbation and following recovery, our data suggest that cytokines generated during the host antiviral response lead to upregulation of the high-affinity IgE receptor, FcεR1, on circulating monocyte/dendritic cell populations, which are the precursors of the antigen presenting cells in the airway mucosa. Upregulation of this receptor would enhance the cells' ability for antigen uptake and processing of aeroallergens for presentation to T_H2 effector cells. If this occurs in subjects presensitized to perennial indoor allergens, which are present at the time of infection, the resultant activation of downstream IgE- FcεR-dependent effector mechanisms in a variety of cell types could contribute significantly to the intensity and duration of ensuing symptoms.[20] Longitudinal studies in infants and young children during respiratory viral infections will be required to determine whether this mechanism contributes to the epidemiologic observations linking early-life viral infections and early allergic sensitization to asthma later in childhood.

CAN THE NEXUS AMONG VIRAL RESPIRATORY INFECTIONS, ATOPY, AND ASTHMA BE BROKEN?

A key component in developing therapeutic options to prevent the development of asthma is to be able to identify those children who are truly at high risk.[4] Clearly every child who develops a wLRI in early life is not at risk of childhood asthma. As is clear from the majority of the epidemiologic literature, wheeze is used both as an indication of LRI and as an indication of developing asthma. However, as outlined, wheeze is a physiological phenomenon and gives no clue to the cause of the wheeze. Thus, wheeze is at best a fuzzy phenotype. Efforts to quantify the LRI better may help to identify high-risk children. We have identified early and persistent allergic sensitization as 1 factor that increases risk of children with wLRI developing asthma.[4,7] The severity of the wLRI may also help identify those at risk; children hospitalized for RSV bronchiolitis have traditionally been considered to be at increased risk of recurrent respiratory problems, including asthma. However, as outlined, the majority of these children do not end up with persistent asthma. Again, lack in the precision of defining the type of asthma may contribute to confusion. Many children diagnosed and treated for asthma by family practitioners and

pediatricians in early to mid childhood "grow out of" their asthma. Asthma associated with atopy is more likely to persist into adulthood,[4] and preventing this type of asthma is likely to have the biggest impact on decreasing the community burden of asthma.

A number of existing therapeutic strategies have the potential to break the nexus between viral respiratory infections and persistent asthma (see review[5]). One uncontrolled trial of palivizumab suggested that preventing or reducing the severity of RSV bronchiolitis in infancy reduces recurrent wheezing in childhood. Similarly, treating infants who wheeze with HRV LRI with prednisolone appeared to reduce the rate of subsequent recurrent wheezing. These studies should be regarded as providing proof of concept that preventing or reducing the severity of viral LRI may decrease the risk of subsequent asthma; however, more specifically targeted therapies are likely to be required. Vaccines against the common respiratory viruses, especially RSV and HRV, are problematic and unlikely to be available in the near future. Potential therapeutic targets that may limit the spread of respiratory viruses from the upper to lower airways and/or reduce the severity of the LRI could include efforts to improve mucosal immunity by using the oral administration of bacterial products, local administration of viral antigens to the upper airway to induce allergen-specific secretory IgA antibodies, efforts to overcome the local deficiency of type I and type III IFNs in the upper and lower airways, or interventions designed to inhibit atopy-related effectors mechanisms from participating in the host response to respiratory viral infection. Alternatively, efforts to prevent allergic sensitization in early life may also decrease the risk of subsequent asthma imposed by early-life viral respiratory infections.

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