

Original Article

The Association of Early Life Viral Respiratory Illness and Atopy on Asthma in Children: Systematic Review and Meta-Analysis

Vikas Wadhwa, MBBS^a, Caroline J. Lodge, PhD^{a,b}, Shyamali C. Dharmage, PhD^{a,b}, Raisa Cassim, MSc^{a,b}, Peter D. Sly, DSc^c, and Melissa Anne Russell, PhD^{a,b} Parkville, VIC, Australia; and South Brisbane, QLD, Australia

What is already known about this topic? Early life viral respiratory illness is commonly associated with wheezing.

What does this article add to our knowledge? There is a difference between atopic and nonatopic children in the association between viral respiratory illness and risk for subsequent asthma/wheeze; however, available evidence is not conclusive. Further studies that consider early life atopy with human rhinovirus infection are warranted.

How does this study impact current management guidelines? The study results recommend further research into the identification of patterns of allergic sensitization that in association with early life viral illness pose the greatest risk for developing asthma.

BACKGROUND: The interaction between early life viral respiratory illness and atopy in the genesis of asthma has been widely discussed in the literature as the “two-hit hypothesis.” **OBJECTIVE:** To synthesize evidence regarding the association of childhood viral respiratory illness and atopy in the development of persistent wheezing and asthma. **METHODS:** A systematic review was performed, according to Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) guidelines. Human studies investigating early life associations between atopy and viral respiratory illness with outcomes of asthma and wheezing were included. Meta-analysis was performed to investigate the association of viral illness across atopic and nonatopic groups. Subgroup analysis was undertaken to investigate potential effect modification of age at outcome. **RESULTS:** Nine cohort studies were included, with data available for meta-analysis in 4 birth cohort studies. There was a stronger association of viral respiratory illness with persistent asthma/wheeze in atopic (odds ratio [OR], 4.02; 95% CI, 1.46–11.06) compared with nonatopic (OR, 2.32; 95% CI, 1.22–4.40) individuals; however, the evidence for this was limited. In 3

studies amenable to subanalysis based on outcome age, a stronger effect was observed up to 7 years for those with atopy (OR, 7.27; 95% CI 4.65–11.36) compared with those without atopy (OR, 3.19; 95% CI, 2.09–4.87).

CONCLUSIONS: There was a stronger association between viral respiratory illness and asthma/wheeze outcomes in individuals with atopy as compared with those without atopy. When outcomes were considered at younger ages, a greater differential effect was observed. Within the limitations of the few available studies however, definite conclusions cannot be made. There was also insufficient evidence for differential effects of early versus late atopy. Further research, in particular regarding virus type, timing of atopy, and atopic phenotype, would contribute to untangling this complex association. © 2020 American Academy of Allergy, Asthma & Immunology (J Allergy Clin Immunol Pract 2020;■:■–■)

Key words: Aeroallergen; Asthma; Atopy; Epidemiology; Viral illness

INTRODUCTION

Asthma is the most common chronic disease in childhood, with large and varying global prevalence, affecting up to 14% of all children.^{1–4} It has a diverse and heterogeneous phenotypic expression,⁵ related to airway inflammation, reversible obstruction, and hyperresponsiveness. Multiple interconnected gene-environmental and epigenetic risk factors are implicated in its causation.^{6–11} The “two-hit hypothesis” suggests that atopy and viral respiratory infections interact in asthma development.^{4,12} These factors are both common and have opportunities to exert effects on immature and developing respiratory systems.^{13–15}

Different mechanisms have been proposed to explain the hypothesized synergy between viral respiratory illness and atopy in asthma risk.^{13,15–17} *In vitro* studies of human respiratory

^aAllergy and Lung Health Unit, Centre for Epidemiology and Biostatistics, School of Population and Global Health, University of Melbourne, Parkville, VIC, Australia

^bMurdoch Children’s Research Institute, Parkville, VIC, Australia

^cCentre for Children’s Health Research, South Brisbane, QLD, Australia

Conflicts of interest: The authors declare that they have no relevant conflicts of interest.

Received for publication May 17, 2019; revised February 22, 2020; accepted for publication March 18, 2020.

Available online ■■

Corresponding author: Melissa Anne Russell, PhD, Centre for Epidemiology and Biostatistics, School of Population and Global Health, University of Melbourne, Level 3 207 Bouverie St, Parkville, VIC 3010, Australia. E-mail: melissar@unimelb.edu.au.

2213-2198

© 2020 American Academy of Allergy, Asthma & Immunology

<https://doi.org/10.1016/j.jaip.2020.03.032>

Abbreviations used

COAST- Childhood Origins of Asthma

HRV- Human rhinovirus

OR- Odds ratio

RSV- Respiratory syncytial virus

epithelial cells confirm disruption to airway barrier function and greater paracellular permeability by rhinovirus infection.¹⁸ Defects in innate immunity may further predispose some individuals to altered responses to viral infections, thereby facilitating the genesis of asthma.¹⁹ Differences between respiratory viruses such as respiratory syncytial virus (RSV) and human rhinovirus (HRV) and their relationships with underlying atopy have also been noted.¹³ Evidence that supports greater susceptibility to asthma in atopic children has been predominantly focused on IgE and T_H2 responses; however, there is likely to be greater complexity in immune mechanisms, which may determine eventual asthma phenotypes.^{20,21}

We aimed to synthesize the evidence regarding the hypothesis that for children with atopy, viral respiratory illness in early life (age up to 5 years) results in increased risk of subsequent wheezing, asthma, and lung function impairment, compared with children without atopy. Secondary aims were to determine whether these relationships were influenced by (1) age of confirmed atopy, (2) age at outcome measurement, and (3) frequency of early life respiratory illness.

METHODS**Search strategy**

PubMed and Embase (Elsevier) databases were searched for original articles that explored associations between exposures to viral respiratory illness and atopy with outcomes of asthma and wheezing. Searches were refreshed until the final date of July 16, 2018. Key terms included “asthma,” “wheezing,” “allergic sensitization,” “atopy,” “virus,” and “illness” (see this article’s Online Repository at www.jaci-inpractice.org). Cross-checking reference lists of published articles was undertaken to ensure all relevant articles were considered.

Article screening and identification

Articles were initially screened by title and abstract and removed if inclusion criteria detailed below were not satisfied. Full-text articles were retrieved and reviewed to determine appropriateness for inclusion. Authors were contacted for further information where this information was not complete or clear.^{14,22-25} Screening of articles was undertaken by 2 independent researchers (V.W. and R.C.). Any disagreements were resolved by a third and independent investigator (M.R.).

Inclusion and exclusion criteria

All English language studies that contained data regarding (1) early life viral respiratory illness in atopic and nonatopic children, or (2) early life sensitization in children with and without a history of viral respiratory illness, or (3) included an interaction term between viral respiratory illness and atopy for subsequent outcomes of persistent wheezing, asthma, or lung function in humans were included. Studies including only subsets of individuals, that is, no comparison between atopic and nonatopic children, were excluded. Atopy was defined by the presence of allergen specific IgE or positive

skin prick testing results. Age at outcome was not a restriction; however, exposures during early childhood were necessary for inclusion in the systematic review. Viral respiratory illness was defined as the presence of compatible viral respiratory symptoms, including fever or wheezing, and assessed by questionnaires or through viral detection methods including culture and PCR.

Data extraction

Data extracted included authors, year of publication, country, study population, type of study (cohort, cross-sectional, or case-control), timing of exposures, method of determining exposure to infection (history, symptoms, isolation of viruses by culture, immunofluorescence, or PCR), definition of allergy exposure (total or specific IgE, skin prick testing), method for determining outcome, and age at outcome assessment. Confounding variables, measures of association (odds ratios [ORs], risk ratios, and hazard ratios) and *P* values for interaction, and authors’ conclusions were extracted.

Quality assessment

All included articles were assessed for quality by using the Newcastle-Ottawa Scale.²⁶ The Newcastle-Ottawa Scale considers selection, comparability, and exposure, with a maximum of 9 points awarded for cohort and case-control studies.

Analysis

A random-effects meta-analysis of the data provided in the articles was performed to evaluate the association of viral respiratory illness across atopic and nonatopic groups separately. Studies reporting ORs and 95% CIs, or where data could be used to calculate these, were included. Subgroup analyses were planned to assess the modifying effect of (1) timing of atopy (<2 y vs ≥2 y), (2) age at outcome (7 y vs ≥7 y), and (3) frequency of viral illness. All data were statistically analyzed using Stata 14.2 (Statacorp, College station, Texas).

RESULTS**Results of electronic database searches**

Database searches revealed a total of 1935 records. After removal of 281 duplicates, 1654 records were screened according to their title and abstract, and 1565 of these were removed as not meeting inclusion criteria for the systematic review. Full text was obtained for the remaining 89 articles. Of these, 80 articles did not meet inclusion criteria and a total of 9 studies were retained for analysis (Figure 1).

Description of studies included in the systematic review

Study designs and locations. From the 9 included studies, there were 8 birth cohort studies^{13-17,27-29} representing 6 populations, with 4 studies analyzing the same cohorts (2 studies for each cohort) at separate time points (see Table E1 in this article’s Online Repository at www.jaci-inpractice.org).^{13-15,17} Of these, 5 were high-risk birth cohort studies,¹³⁻¹⁷ 2 were population based,^{27,28} and 1 was hospital based.²⁹ The remaining study was a prospective cohort study with recruitment in infancy.³⁰

Populations. Kusel et al^{15,17} published results from a high-risk Australian birth cohort of 198 children aged 5 years in 2007,¹⁵ with follow-up at age 10 years in 2012.¹⁷ The Childhood Origins of Asthma (COAST) from the United States was also a high-risk birth cohort of 259 children followed up at age 6

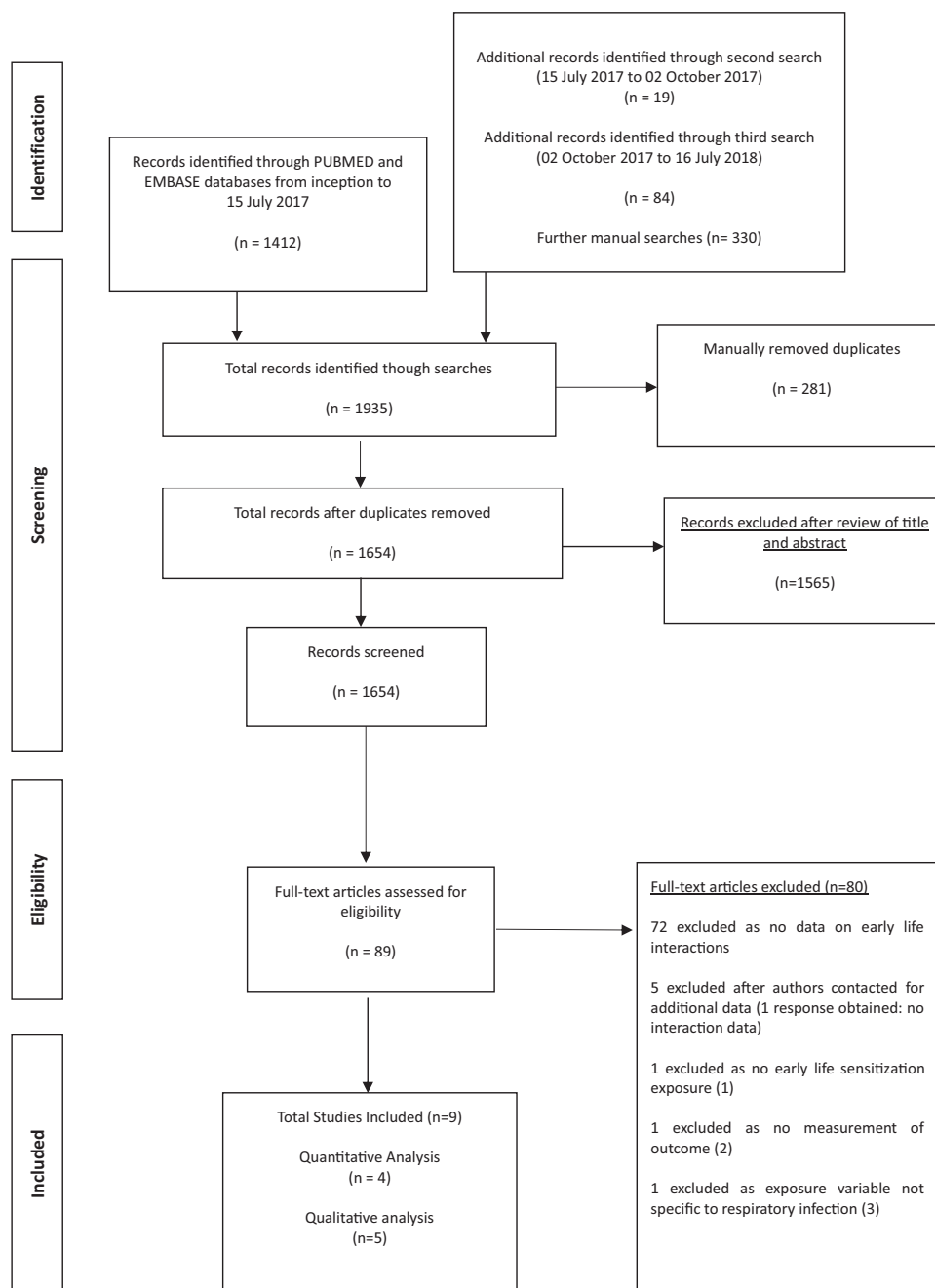


FIGURE 1. PRISMA flow diagram. *PRISMA*, Preferred Reporting Items for Systematic reviews and Meta-Analyses.

years¹³ and again at age 13 years.¹⁴ Studies with an interaction effect published were a review by Oddy et al²⁷ of a Western Australian pregnancy prospective birth cohort with 2602 individuals and the Oslo birth cohort study by Nafstad et al²⁸ of 2540 individuals. The study by Henderson et al²⁹ comprising 150 hospitalized individuals from the longitudinal birth cohort of Avon Longitudinal Study of Parents And Children from the United Kingdom was included, as was the study by Bønnelykke et al¹⁶ of 313 high-risk birth cohort from the Copenhagen Prospective Studies of Asthma in Childhood (2000) from Denmark. A prospective infant cohort study by Lukkari-

et al³⁰ from Finland comprised 127 wheezing infants enrolled at a median age of 11 months and followed for 7 years.

Four birth cohort study publications, each representing a distinct population, were eligible for inclusion in the meta-analysis.^{15,16,27,28} One study included a hospital-based population and was excluded from the meta-analysis, but retained for qualitative analysis.²⁹ The remaining 4 cohort studies did not have adequate data in a format to be included in the meta-analysis but were retained for qualitative assessment.^{13,14,17,30} Within the studies retained only for a qualitative review, 1 had data included in the meta-analysis from a previous article.¹⁷

Measures of exposures

Viral respiratory illness. Viral respiratory illness was assessed within the first year of life in most studies.^{13-15,17,27-29}

In the study by Oddy et al,²⁷ exposure was documented as wheezy lower respiratory tract illness, whereas in the article by Nafstad et al,²⁸ a history of viral illness was recorded, both using questionnaires. In the remaining studies, evidence of virus infection with virological data was obtained, with 2 of these cohort studies^{16,30} measuring viral respiratory infections between age 1 and 3 years. Detection methods of viral respiratory infection however varied across the studies.

PCR. Viral infection was confirmed by PCR of nasopharyngeal aspirates in 6 cohort studies,^{13-17,30} but not in 3 cohort studies.²⁷⁻²⁹ PCR testing was used to detect HRV and RSV and other pathogens. Differences in timing and sampling of respiratory secretions were observed across studies that involved PCR (see this article's Online Repository at www.jaci-inpractice.org).

Immunofluorescence and culture. In the Avon Longitudinal Study of Parents And Children cohort, nasopharyngeal aspirates were taken during hospitalization for bronchiolitis symptoms although the timing was not specified.²⁹ Immunofluorescence or culture of aspirates was used for detection of RSV infection only.²⁹

Evidence for exposure was determined through questionnaire and clinical data in the Oslo and the Western Australia Pregnancy Cohort studies.^{27,28} The Western Australian study also considered the number of respiratory illnesses and the presence of wheezing.²⁷ Symptom data on questionnaires included history of lower and upper respiratory tract infections in early life and also at multiple time intervals.

Atopy. Measurement for atopy was uniform across the 5 cohort studies included in the meta-analysis^{15,16,27-29} and 2 further cohort studies in the qualitative analysis,^{13,17} all using positive skin prick testing. The timing of skin prick testing however ranged from 6 months up to 10 years. In the meta-analysis, only the Kusel et al^{15,17} studies considered skin prick testing by the age of 2 years, with the remaining studies evaluating responses at ages 5 to 10 years. In addition, in this study a skin prick test wheal of diameter 2 mm at ages 6 months and 2 years was considered a positive result, whereas other studies considered a wheal diameter of 3 mm or more to at least 1 allergen to be indicative of sensitization.¹⁵ The remaining 2 cohort studies^{14,30} used specific serum IgE as measures of sensitization. Two cohort studies used both skin prick testing and specific serum IgE at ages 5 to 6 years.^{13,16} Studies by Kusel et al^{15,17} separated allergic sensitization into early (before age 2 years) and delayed (after age 2 years). The COAST studies included within the qualitative analysis had measurement of atopy using specific IgE before the age of 2 years and with skin prick testing at age 5 years.

Measures of outcomes. In all studies, asthma was the main outcome measure and in birth cohort analyses, doctor diagnosis was the definition used.^{13-17,27-30} Ages at assessment varied from less than 5 years¹⁵ in an Australian birth cohort up to 10 years in the Oslo cohort.²⁸ There was an absence of lung function data within the included studies.

The COAST cohort studies assessed outcome at multiple age points (6, 8, 11, and 13 years) for current asthma using

either doctor diagnosis, use of asthma medications, or step-up management plans.¹⁴ In addition, wheezy respiratory illness in the first 3 years of life was defined as either doctor diagnosed or symptoms for which bronchodilator or symptom controller therapy was prescribed. Wheezing was included as an additional outcome in the Avon Longitudinal Study of Parents And Children cohort through reported symptoms.²⁹ The Copenhagen Prospective Studies of Asthma in Childhood 2000 study required favorable response to 3 months of inhaled steroids and relapse on withdrawal to support a doctor diagnosis of asthma.¹⁶

In the Lukkarinen et al³⁰ study, current asthma outcome at age 8 years was defined through either (1) doctor diagnosis and need for asthma treatment, (2) use of oral corticosteroids or bronchodilators, or (3) demonstration of reversibility or hyper-reactivity on spirometry.

Confounding variables. There was variability in confounders included in the studies (see Table E1). Consistently, passive smoke and parental asthma/atopy were included. Breast-feeding was considered in 7 studies^{13-15,17,28-30} and older siblings in 5 studies.^{13-15,17,27} Sex was controlled for in 5 birth cohort studies^{15,17,27-29} and maternal education in 4 studies.^{16,17,27,28} Other variables recognized as confounders across some studies included gestational age,²⁷ month or season of birth,^{13,14,29} day care utilization,^{13-15,17} and exposure to pets.^{13-15,17}

Quality assessment of included studies. From a possible maximum of 9 points in the Newcastle-Ottawa Scale, 8 points, indicating good quality, were recorded for 5 cohort studies^{13-15,17,30} that represented 3 distinct populations (see Table E2 in this article's Online Repository at www.jaci-inpractice.org). Seven points were recorded for 2 studies,^{16,27} representing 2 distinct populations. Two further studies scored 6 and 5 points, respectively, each representing a distinct population.^{28,29}

Findings of studies included in the systematic review but not the meta-analysis. Four cohort studies (3 birth, 1 infancy)^{13,14,17,30} that represented 3 distinct populations could not be included in the meta-analysis because data were not provided in a format that permitted calculation of effect size. Another cohort study could not be included in the meta-analysis because the population was infants hospitalized with RSV bronchiolitis.^{28,29}

The COAST cohort studies found the greatest effect on asthma outcomes observed with HRV infections measured in the first year of life (and with respiratory illness) and atopy.^{13,14} They demonstrated an interaction effect when the outcome was evaluated at age 6 years with an OR of 45, when in comparison, the OR for asthma diagnosis was 3.9 for atopy and 7.8 for HRV alone.¹⁴ However, by the age of 13 years, the OR for asthma with atopy below age 3 years was 2.7, for HRV the OR was 3.0, and combined the OR was 7.9. Data regarding the precision of this estimate were not available.

Kusel et al¹⁷ in 2012 investigated the effects of atopy and febrile, wheezy viral lower respiratory illnesses in a high-risk birth cohort, previously analyzed at age 5 years in 2007. They found that at age 10 years, for the effect of viral respiratory illness on asthma, the risk ratio was 4.92 (95% CI, 2.59-9.36) with early sensitization compared with children with delayed sensitization

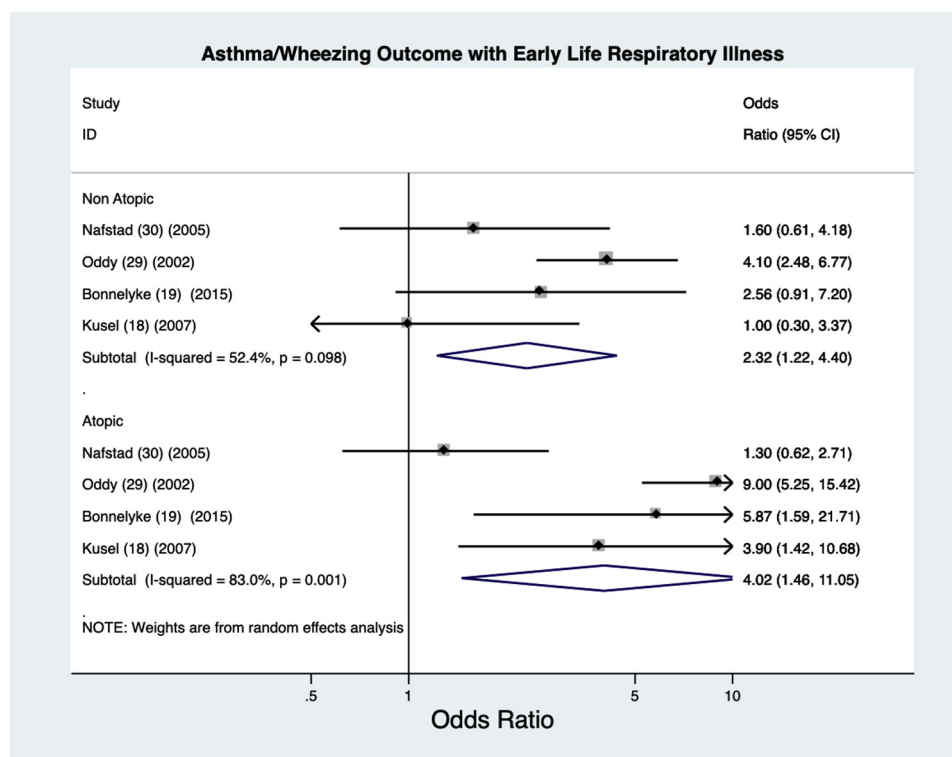


FIGURE 2. Meta-analysis of the association of early life respiratory illness with asthma/wheeze outcome across nonatopic and atopic subjects.

in whom the risk ratio was 1.13 (95% CI, 0.19-6.79); there was significant overlap of CIs.

The Lukkarinen et al³⁰ study considered 127 children initially presenting with severe wheezing and followed for 7 years. A greater proportion of children with both HRV infection, measured at a mean age of 11 months, and atopy was diagnosed with current asthma (59%) in comparison to those with either factor (21%) or none (25%). Data regarding the precision of this estimate were not available.

Meta-analysis results

Four cohort studies were eligible for inclusion in the meta-analysis.^{15,16,27,28} Results demonstrated a difference in the association of viral respiratory illness with asthma and wheeze outcomes across those with and without atopy, with an observed OR of 4.02 (95% CI, 1.46-11.05; $I^2 = 83\%$) and 2.32 (95% CI, 1.22-4.40; $I^2 = 52\%$), respectively (Figure 2). However, it should be noted that the evidence for this conclusion is limited because of the small number of studies and participants. Considered individually, none of the cohort studies included in the meta-analysis demonstrated an interaction between viral respiratory illness and atopy for the outcome of wheeze and asthma. In those studies in which viral wheezing illness was measured in the first year of life, individuals there was a slight but not statistically significant greater risk for asthma in atopics than in nonatopic individuals.^{13,27}

Meta-analysis substudies

Age at measurement of outcome. Outcome was measured at or below age 7 years in 3 of the 4 cohort studies^{15,16,27}

and above 7 years in a single cohort study.²⁸ In atopic children with symptomatic early life viral illness, a moderate increase in risk for asthma and wheeze was observed (OR, 7.09; 95% CI, 4.36-11.53; $I^2 = 8\%$) in comparison to the nonatopic group (OR, 2.54; 95% CI, 1.16-5.56; $I^2 = 57\%$) when the outcome was measured at an age of 7 years or below (Figure 3). In the single study in which the outcome was measured at an age above 7 years, there was minimal difference in risk of asthma with early life exposure to viral illness between atopic (OR, 1.30; 95% CI, 0.62-2.71) and nonatopic children (OR, 1.60; 95% CI, 0.61-4.18).

Frequency of viral respiratory illness. In the 2 included studies, comparison was made between 1 and 0 illness, and 2 or more and 0 respiratory illness, in the first year.^{15,27} Exposure to frequent respiratory illness portended a moderate increase in risk for wheezing and asthma outcome in atopic individuals (OR, 8.81; 95% CI, 5.27-14.71) compared with similarly exposed nonatopic individuals (OR, 3.16; 95% CI, 1.08-9.23) (see Figure E1 in this article's Online Repository at www.jaci-inpractice.org). In those with less frequent illness, there remained an increased risk for wheezing and asthma in atopic individuals (OR, 3.52; 95% CI, 2.11-5.88) compared with nonatopic individuals (OR, 1.68; 95% CI, 0.80-3.51) (see Figure E1).

Age at measurement of atopy. The Kusel et al¹⁵ study was the only study in the meta-analysis in which atopy was measured before the age of 2 years. It demonstrated a greater effect on asthma/wheeze outcome from early life respiratory illness in atopic individuals compared with nonatopic

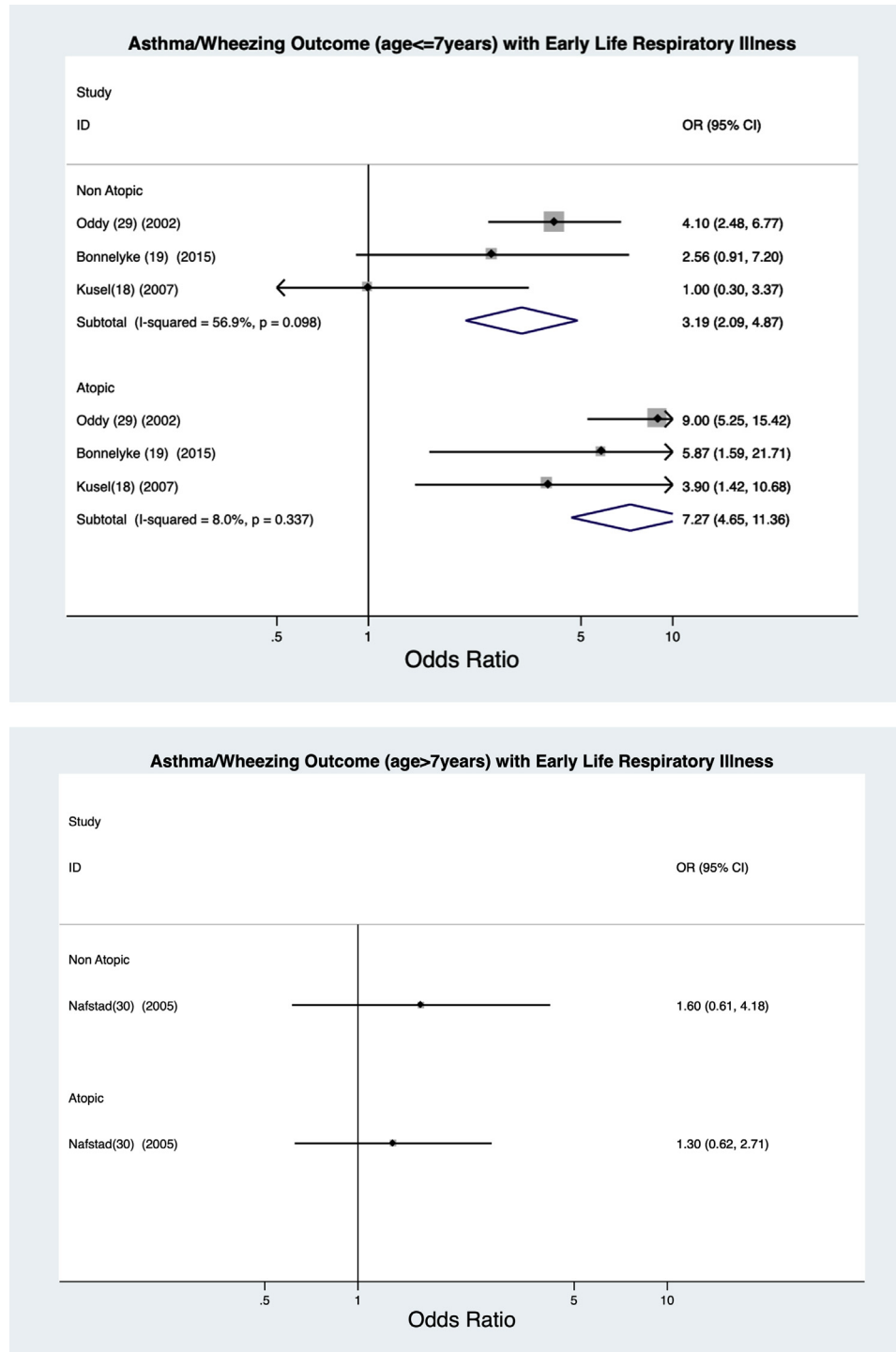


FIGURE 3. Meta-analysis of the association of early life respiratory illness with asthma/wheeze outcome across nonatopic and atopic subjects by age of outcome measurement.

individuals, with an OR of 3.90 (95% CI, 1.42-10.68) and 1.00 (95% CI, 0.30-3.37), respectively¹⁵ (Figure 4). In the other 3 studies, atopy was measured after the age of 2 years.^{16,27,28} In these articles, a smaller differential association between those with and without atopy was seen, with an observed OR of 4.06 (95% CI, 1.04-15.87; $I^2 = 89\%$) in

children with atopy and 2.91 (95% CI, 1.65-5.15; $I^2 = 36\%$) in children without atopy. These 3 studies^{16,27,28} were also different from the Kusel et al article in that measurements of exposure were at a similar time to outcome, thereby further limiting conclusions to an effect association rather than causation (Figure 4). The Nafstad et al article was

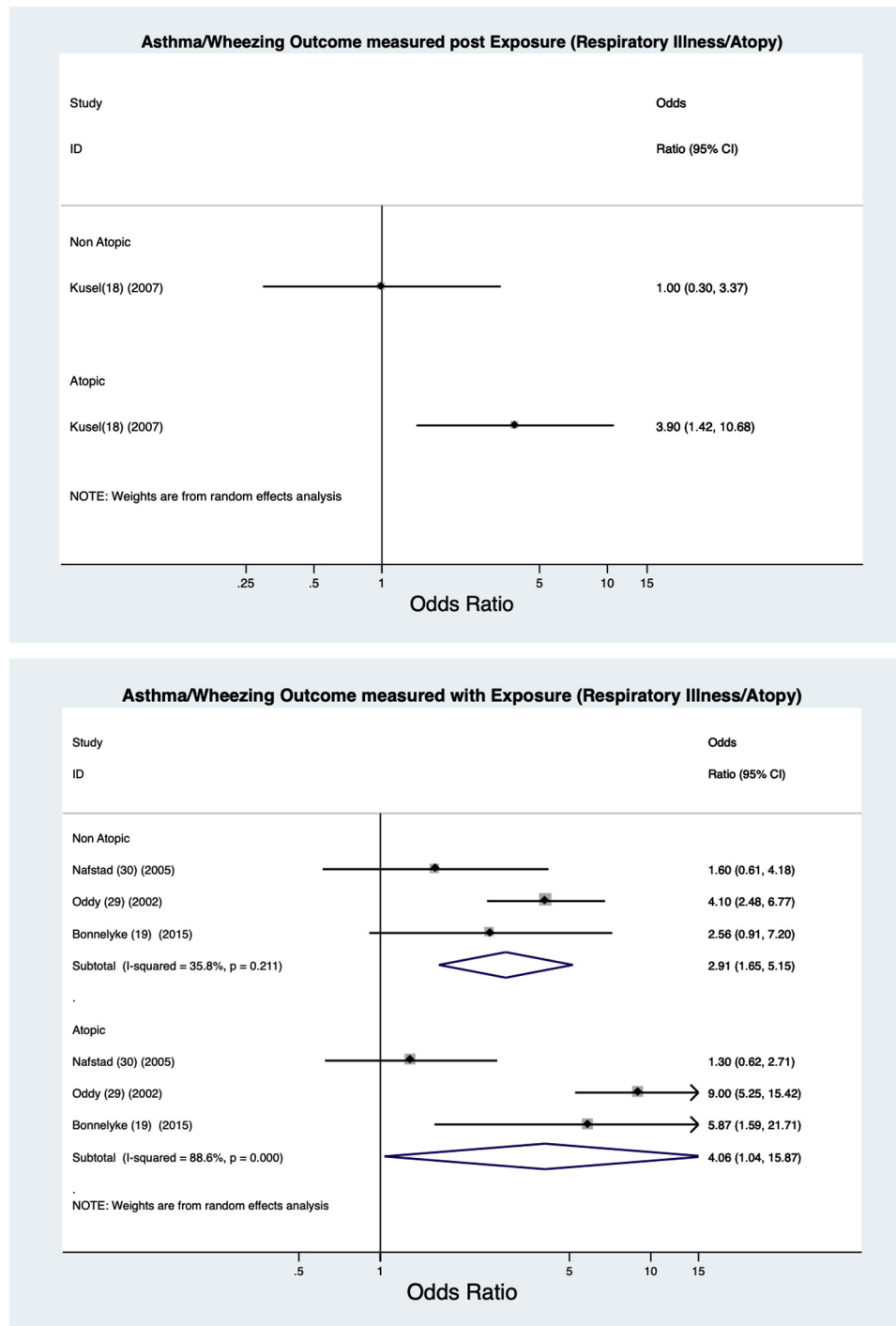


FIGURE 4. Asthma/wheezing in nonatopic and atopic subjects by relative timing of exposure and outcome measurements.

notable for measurement of atopy and asthma outcome much later at age 10 years. When removed from analysis, a greater effect on asthma/wheeze outcome was observed in atopic individuals compared with nonatopic individuals, with an observed OR of 8.46 (95% CI, 5.14-13.92; $I^2 = 0\%$) and 3.75 (95% CI, 2.39-5.89; $I^2 = 0\%$), respectively (Figure 5).

DISCUSSION

This is the first systematic review that considers the association of viral respiratory illnesses with and without atopy in children, in an attempt to determine effects on the outcomes of wheezing and asthma. In the 4 cohort studies included in the qualitative review,^{13,14,17,30} greater effects with the combination of both

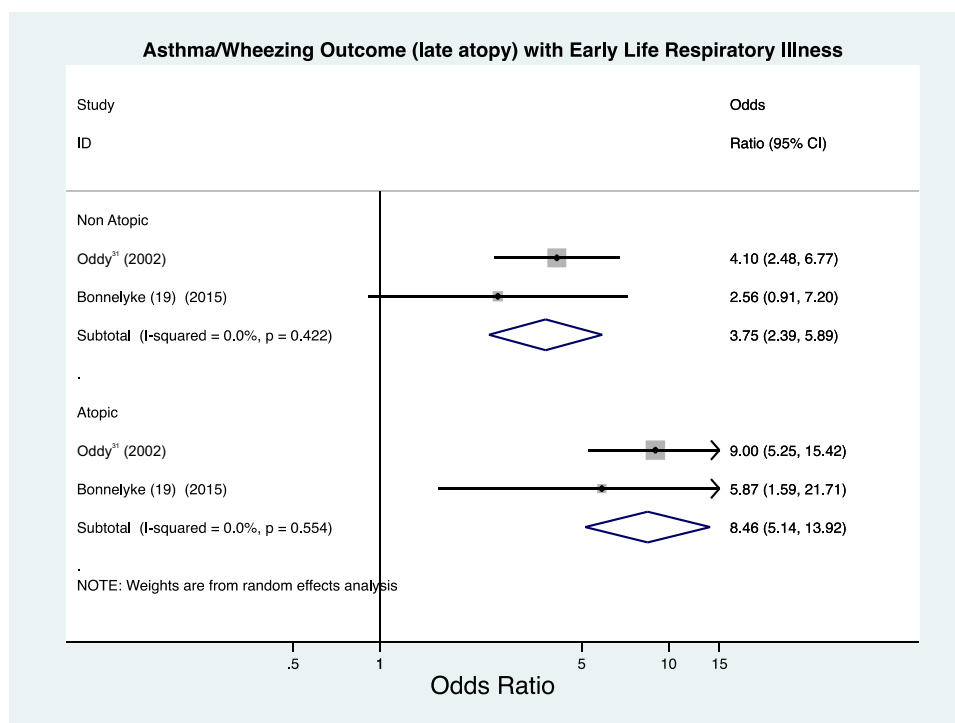


FIGURE 5. Association of early life respiratory illness and late atopy with asthma/wheeze outcome across nonatopic and atopic subjects.

factors were observed. Within the meta-analysis, there was some difference in the association of viral respiratory illnesses with asthma and wheeze across atopic and nonatopic populations when articles were considered in the meta-analysis. When results were limited to studies assessing outcome up to 7 years and when measurements of early age atopy were compared with later age, differences across the 2 populations were greater. Available evidence however limits scope for strong conclusions.

An immature and developing respiratory and immune system is considered highly vulnerable to viral infections and may explain the observed differences when outcome is measured at an early age. However, one of the major challenges is that in the very young it may be difficult to establish a confirmatory diagnosis of asthma and hence, interactive effects between the 2 exposures may be missed when outcome is assessed prior to when a definitive diagnosis can be made. An additional factor to consider is that the interaction between atopy and respiratory viral illness may be associated with certain wheeze phenotypes but not others.³¹⁻³³ In particular, it may be that the interaction between the 2 factors may be more strongly associated with prolonged early wheeze and/or intermediate-onset wheeze phenotypes; these phenotypes have a higher prevalence around age 5 years and then become less common.³²

Studies of high-risk birth cohorts suggest potential differences between viral types, notably outlining the greater impact of specific HRV subtypes over RSV.^{13,14} Virus type has however not been consistently defined as a differentiating factor, particularly in the absence of comparable determinants of severity, frequency, and duration of infection and precise timing of

infection. Inability to detect certain viruses, particularly in earlier studies not using PCR, may be a further limitation that clouds differences between potential pathogens. It is possible that distinct pathways exist for different viruses and eventual phenotypes are determined by the intersection of multiple variables including host immune factors, microbiomes, and genetic susceptibility.

Notably only 2 studies in the meta-analysis, representing 1 population, measured atopy at age less than 1 year.^{15,17} Although low power limits the conclusions that can be made from a single population, there was some indication of a synergistic effect within these articles.^{15,17} Although previous research has indicated that infants born into allergic families do not usually have high-level specific IgE antibodies to aeroallergens before the age of 2 years,³⁴ early and rapidly rising specific IgE responses have been associated with persistence of sensitization at age 5 years.³⁵ Further complicating this are dissimilarities in the way atopy is diagnosed.^{36,37} Differential sensitization patterns may exert dissimilar effects on asthma risk, severity, and persistence,^{3,14,38,39} with the greatest impact currently considered to be wielded by early multiple and aeroallergen sensitization.³⁹ In another birth cohort study, mold rather than other aeroallergen sensitization was observed to have greater association with asthma diagnosis at age 7 years in the presence of high numbers (6 or more) of respiratory infections.⁴⁰ Different atopic phenotypes may therefore have potential to interact distinctively with early life viral respiratory infections in defining future asthma risk.

Limitations of this systematic review include the small number of studies and sample sizes, particularly when

considering studies with similar ages at exposure and outcome measurements. Of note is absence of lung function data within the included studies. PCR is acknowledged as the more sensitive measure of viral detection, and it should be noted that most, but not all, studies included in this review used this method. Failure to detect definite interactions between those with and without atopy may also be influenced in terms of differences in sex, race, wheal size used in the SPT, and airway size, factors unable to be explored within the current meta-analysis. Hence, conclusions regarding this important consideration are limited.

It is pertinent to note that the studies included in our meta-analysis predominantly comprise high-risk groups. Although overall there was limited evidence to support the two-hit hypothesis, there was some indication from individual studies that the hypothesis holds when atopy develops early in life, with HRV infection and predominantly when asthma is assessed at younger ages. Our article highlights the importance of further research to determine whether this association is consistent across other birth cohort studies and whether stronger focus on the atopy component of the two-hit hypothesis and viral type may shed further light as to unique susceptibilities and true impacts on asthma and lung function.

CONCLUSIONS

In this systematic review and meta-analysis, we observed a differential effect of viral respiratory illnesses across nonatopic and atopic groups, particularly when outcome analyses were limited to studies conducted at younger ages. There was also a differential effect within individual studies that considered atopy early in life. Within limitations of the small number of studies available for inclusion in this systematic review and meta-analysis we found some but insufficient evidence to conclusively confirm the long-held acceptance of interactions between atopy and early life viral respiratory illnesses in the causation of asthma and persistent wheezing. Implications for public health are that protection of the developing respiratory system in at-risk groups through preventative measures such as immunization, antiviral therapy, and potentially interventions to enhance T_H1 immunity may prove crucial. A targeted and multifaceted approach to develop innate immunity and minimize risks to noxious agents should be our intent.

REFERENCES

- Bosken CH, Hunt WC, Lambert WE, Samet JM. A parental history of asthma is a risk factor for wheezing and nonwheezing respiratory illnesses in infants younger than 18 months of age. *Am J Respir Crit Care Med* 2000;161:1810-5.
- Subbarao P, Mandhane PJ, Sears MR. Asthma: epidemiology, etiology and risk factors. *CMAJ* 2009;181:E181-90.
- Jackson DJ, Gern JE, Lemanske RF. The contributions of allergic sensitization and respiratory pathogens to asthma inception. *J Allergy Clin Immunol* 2016;137:659-65.
- Gern JE. Virus/allergen interaction in asthma exacerbation. *Ann Am Thorac Soc* 2015;12:S137-43.
- Holt PG, Sly PD. Interaction between adaptive and innate immune pathways in the pathogenesis of atopic asthma: operation of a lung/bone marrow axis. *Chest* 2011;139:1165-71.
- Carroll KN, Gebretsadik T, Minton P, Woodward K, Liu Z, Miller EK, et al. Influence of maternal asthma on the cause and severity of infant acute respiratory tract infections. *J Allergy Clin Immunol* 2012;129:1236-42.
- Frick OL, German DF, Mills J. Development of allergy in children, I: association with virus infections. *J Allergy Clin Immunol* 1979;63:228-41.
- Kurukulaaratchy RJ, Fenn M, Matthews S, Arshad SH. Characterisation of atopic and non-atopic wheeze in 10 year old children. *Thorax* 2004;59:563-8.
- Lemanske RF Jr. Issues in understanding pediatric asthma: epidemiology and genetics. *J Allergy Clin Immunol* 2002;109:S521-4.
- Belsky DW, Sears MR. The potential to predict the course of childhood asthma. *Expert Rev Respir Med* 2014;8:137-41.
- Sly PD, Kusel M, Holt PG. Do early-life viral infections cause asthma? *J Allergy Clin Immunol* 2010;125:1202-5.
- Rosenthal LA, Avila PC, Heymann PW, Martin RJ, Miller EK, Papadopoulos NG, et al. Viral respiratory tract infections and asthma: the course ahead. *J Allergy Clin Immunol* 2010;125:1212-7.
- 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.
- Rubner FJ, Jackson DJ, Evans MD, Gangnon RE, Tisler CJ, Pappas TE, et al. Early life rhinovirus wheezing, allergic sensitization, and asthma risk at adolescence. *J Allergy Clin Immunol* 2017;139:501-7.
- Kusel MM, de Klerk NH, Keadze 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.
- Bønnelykke K, Vissing NH, Sevelsted A, Johnston SL, Bisgaard H. Association between respiratory infections in early life and later asthma is independent of virus type. *J Allergy Clin Immunol* 2015;136:81-86.e4.
- Kusel MM, Keadze T, Johnston SL, Holt PG, Sly PD. Febrile respiratory illnesses in infancy and atopy are risk factors for persistent asthma and wheeze. *Eur Respir J* 2012;39:876-82.
- Sajjan U, Wang Q, Zhao Y, Gruenert DC, Hershenson MB. Rhinovirus disrupts the barrier function of polarized airway epithelial cells. *Am J Respir Crit Care Med* 2008;178:1271-81.
- 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.
- Hollams EM, Devereil M, Serralha M, Suriyaarachchi D, Parsons F, Zhang G, et al. Elucidation of asthma phenotypes in atopic teenagers through parallel immunophenotypic and clinical profiling. *J Allergy Clin Immunol* 2009;124:463-70. 470.e1-470.e16.
- Smart JM, Kemp AS. Increased Th1 and Th2 allergen-induced cytokine responses in children with atopic disease. *Clin Exp Allergy* 2002;32:796-802.
- Besednjak-Kocijancic L. Is adenovirus an important risk factor for childhood asthma? *Allergy* 2014;69:547-8.
- Burr ML, Dunstan FDJ, Hand S, Jones KP, Rolf S. The natural history of wheeze from birth to adult life: a cohort study. *Thorax* 2010;65:A51-2.
- van Meel ER, den Dekker HT, Elbert NJ, Jansen PW, Moll HA, Reiss IK, et al. A population-based prospective cohort study examining the influence of early-life respiratory tract infections on school-age lung function and asthma. *Thorax* 2018;73:167-73.
- Frassanito A, Nenna R, Cangiano G, Scagnolari C, Petrarca L, Calicchia I, et al. Asthma after bronchiolitis: five years of follow-up. *Eur Respir J* 2018;52:PA4646.
- Wells GA, Shea B, O'Connell D, Peterson J, Welch V, Losos M, et al. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomized studies in meta-analyses. Available from: http://www.ohri.ca/programs/clinical_epidemiology/oxford.htm. Accessed July 1, 2018.
- Oddy WH, de Klerk NH, Sly PD, Holt PG. The effects of respiratory infections, atopy, and breastfeeding on childhood asthma. *Eur Respir J* 2002;19:899-905.
- Nafstad P, Brunekreef B, Skrandal A, Nystad W. Early respiratory infections, asthma, and allergy: 10-year follow-up of the Oslo Birth Cohort. *Pediatrics* 2005;116:e255-62.
- Henderson J, Hilliard TN, Sheriff 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.
- Lukkarinen M, Koistinen A, Turunen R, Lehtinen P, Vuorinen T, Jartti T. Rhinovirus-induced first wheezing episode predicts atopic but not nonatopic asthma at school age. *J Allergy Clin Immunol* 2017;140:988-95.
- Savenije OE, Granell R, Caudri D, Koppelman GH, Smit HA, Wijga A, et al. Comparison of childhood wheezing phenotypes in 2 birth cohorts: ALSPAC and PIAMA. *J Allergy Clin Immunol* 2011;127:1505-1512.e14.
- Lodge CJ, Zaloumis S, Lowe AJ, Gurrin LC, Matheson MC, Axelrad C, et al. Early-life risk factors for childhood wheeze phenotypes in a high-risk birth cohort. *J Pediatr* 2014;164:289-294.e1-294.e2.
- Granell R, Henderson AJ, Sterne JA. Associations of wheezing phenotypes with late asthma outcomes in the Avon Longitudinal Study of Parents and

- Children: a population-based birth cohort. *J Allergy Clin Immunol* 2016;138:1060-1070.e11.
34. Duff AL, Pomeranz ES, Gelber LE, Price GW, Farris H, Hayden FG, et al. Risk factors for acute wheezing in infants and children: viruses, passive smoke, and IgE antibodies to inhalant allergens. *Pediatrics* 1993;92:535-40.
 35. Holt PG, Rowe J, Kusel M, Parsons F, Hollams EM, Bosco A, et al. Toward improved prediction of risk for atopy and asthma among preschoolers: a prospective cohort study. *J Allergy Clin Immunol* 2010;125:653-9:659.e1-659.e7.
 36. Beck AF, Huang B, Kercsma CM, Guilbert TW, McLinden DJ, Lierl MB, et al. Allergen sensitization profiles in a population-based cohort of children hospitalized for asthma. *Ann Am Thoracic Soc* 2015;12:376-84.
 37. Weinmayr G, Genuneit J, Nagel G, Björkstén B, van Hage M, Pflüger A, et al. International variations in associations of allergic markers and diseases in children: ISAAC phase two. *Allergy* 2010;65:766-75.
 38. Shields MD, McGovern V. Advances in the management of childhood asthma. *Curr Paediatr* 2003;13:264-8.
 39. Simpson A, Tan VY, Winn J, Svensen M, Bishop CM, Heckerman DE, et al. Beyond atopy: multiple patterns of sensitization in relation to asthma in a birth cohort study. *Am J Respir Crit Care Med* 2010;181:1200-6.
 40. Perez Ramirez L, Wendroth H, Martin LJ, Pilipenko VV, He H, Kroner J, et al. High number of early respiratory infections in association with allergic sensitization to mold promotes childhood asthma. *J Allergy Clin Immunol* 2018;141:1921-1924.e4.

ONLINE REPOSITORY**DATABASE SEARCH TERMS****PubMed**

(ALL(asthma) OR ALL(lung function) OR ALL(wheeze*) AND ALL(respiratory tract infect*) OR ALL(respiratory illness*) OR ALL(respiratory infect*) AND ALL(sige) OR ALL(two hit hypothesis) OR ALL(sensitization) OR ALL(skin prick) OR ALL(serum ige) OR ALL(sensitisation) OR ALL(atopy) OR ALL(atopic) OR ALL(double hit hypothesis) OR ALL(food hypersensitivity) OR ALL(aeroallergen) OR ALL(food allergen) OR ALL(specific ige) OR ALL(food hypersensitivities))) with filters of human and English studies.

Embase (Elsevier)

(((((respiratory NEAR/2 (infect* OR virus* OR viral OR illness) AND ('infant'/exp OR 'preschool child'/exp OR 'toddler'/exp OR 'childhood disease'/exp)) OR (respiratory NEAR/2 (infect* OR virus* OR viral OR illness) AND (wheeze* OR respiratory) NEAR/6 (infant OR early OR child*))) AND ('food allergy'/exp OR food NEAR/2 (hypersens* OR allerg*) OR 'atopy'/exp OR (atopy:ab,ti OR atopic:ab,ti) OR (aeroallerg* OR 'aero allerg*') OR 'sensitization'/de OR (allerg* OR skin

NEAR/2 sensitization OR ('skin prick' OR 'prick test'/de) OR 'immunoglobulin e'/de OR ('serum ige' OR 'specific ige') OR 'double hit hypothesis' OR 'two hit hypothesis') AND (('asthma'/exp/mj AND 'risk factor'/exp) OR 'asthma'/exp/dm_et OR 'wheezing'/exp OR wheeze*:ab,ti)) AND [english]/lim) AND [humans]/lim.

ANALYSIS OF EXPOSURE TO VIRAL INFECTIONS

In the Copenhagen Prospective Studies of Asthma in Childhood 2000 cohort, sampling of nasopharyngeal and hypopharyngeal secretions was undertaken 3 days after the onset of respiratory symptoms.^{E1} In the COAST cohort studies, nasopharyngeal aspirates were taken at scheduled clinic visits and during symptomatic infection. Sampling was altered to nasal lavage if the respiratory symptom score was 5 or greater, signifying more severe respiratory illness.^{E2} In studies reported by Kusel et al,^{E3} symptoms of respiratory infection (including fever >38°C, runny nose, cough, or wheeze) were followed closely with daily symptom diaries and nasopharyngeal aspirates within 48 hours of reporting. The Lukkarinen et al^{E4} infancy cohort study used PCR of nasopharyngeal aspirates upon presentation to hospital with an acute wheezing illness but the duration of preceding symptoms was not defined.

TABLE E1. Cohort studies included in systematic review

Author, date Country	Population	Exposure: Infection	Allergy/Atopy	Outcome: Wheezing/ asthma	Confounders	Results	Author's conclusion
Oddy et al ^{E5} (2002) Australia	N = 2602 age 0-6 y	Assessed at 1 y - Illness details Assessed at 6 y - Illness/history (upper and lower respiratory illness in first year)	Atopy = SPT at age 6 y (at least 1 wheal $\geq$ 2 mm)	Doctor-diagnosed asthma at age 6	Sex, gestational age, maternal smoking in pregnancy, maternal asthma, maternal age, education, age of solids introduction, family income, childcare/older siblings	Wheezy LRTI in year 1: Increased risk of asthma: OR, 2.03 (nonatopic) OR, 3.39 (atopic) $\geq$ 2 infections in year 1 increased risk of asthma: OR, 4.1 (nonatopic) ($P < .005$) OR, 9.0 (atopic) ($P < .0005$) Atopy-wheezy LRTI interaction $P =$.701 Up to 3 URTIs = negative risk for asthma (adjusted OR, 0.75; 95% CI, 0.54-1.04) 4 = + URTI = positive risk for asthma (adjusted OR, 1.32; 95% CI, 0.92- 1.89; $P = .057$)	Wheezy LRTI in year 1 and atopy independently associated with increased asthma risk <i>but</i> together effects are multiplicative No interaction between atopy and wheezy LRTI
Nafstad et al ^{E6} (2005) Norway	N = 2540 Birth cohort 0- 10 y $\rightarrow$ OSLO birth cohort	Respiratory infection on surveys at 6 and 12 mo Direct: 6/12, 12/12, 4 y questionnaires Indirect: attended childcare >10 h/wk + older siblings	SPT + whether wheal $\geq$ 3 mm assessed at age 10 y	Questions of asthma symptoms and doctor diagnosis at age 10	Sex, parental atopy, family income, ETS, breast- feeding, maternal age at delivery, maternal education, single parenthood	Asthma past 1 y, (+) SPT and LRTI OR, 1.3 (95% CI, 0.6-2.6) Asthma past 1 year, (-) SPT and LRTI OR, 1.6 (95% CI, 0.6-4.1) Current asthma related to LRTI: OR, 2.0 (95% CI, 1.3-3.0) in year 1	No interaction between atopy and viral respiratory infections identified
Bønnelykke et al ^{E1} (2015) Denmark	High- risk birth cohort N = 313 children followed to age 7 y from COPSAC 2000 cohort (n = 411)	Viruses/bacteria 0-3 y NPA/HPA PCR analysis for viruses/ bacteria Physical examination	Specific IgE > 3.5 kU/L or >3 mm SPT wheal defines atopy SPTs and specific IgE measured at age 6 y	Asthma at age 7 diagnosed by physicians	Genotypes, father asthma, mother education, nicotine in hair by age 12 mo	Unadjusted for number of respiratory episodes: HRV: OR for asthma if sensitized = 3.84 (95% CI, 1.30-11.35) vs 1.81 (95% CI, 0.77-4.23) if not sensitized RSV: OR for asthma if sensitized = 3.35 (95% CI, 1.17-9.58) vs 1.98 (95% CI, 0.85-4.61) if not Any virus: OR = 5.87 (95% CI, 1.59- 21.76) if sensitized vs 2.56 (95% CI, 0.91-7.19) if not	No evidence for interaction or effect modification HRV infection ($P = .12$) Any virus ($P = .18$)
Henderson et al ^{E7} (2005) UK	Longitudinal birth cohort of RSV bronchiolitis hospitalized; age <1 y (n = 150) (controls no admission for bronchiolitis) (n = 14,062) ALSPAC	NPA data from public health laboratory records: RSV isolated by immunofluorescence or culture	SPT atopy age 7 y Atopy = $\geq$ 2 mm wheal	12-mo prevalence of wheeze at 2 ages (30- 42 mo and 69-81 mo) Cumulative prevalence of doctor- diagnosed asthma at 91 mo	Birth weight, sex, season of birth, mother's age at delivery, maternal parity/ maternal smoking in pregnancy/maternal asthma or eczema, maternal education, housing, breast-feeding, ETS	RSV + atopy: = OR, 2.2 (95% CI, 0.3-15.6) for persistent wheezing = OR, 3.9 (95% CI, 0.9-17.7) at 69-81 mo/12 for wheezing = OR, 1.2 (95% CI, 0.3-5.5) for asthma at age 91 mo RSV + nonatopy: OR, 0.8 (95% CI, 0.2-3.4) for wheezing OR, 4.1 (95% CI, 1.8-9.6) in 69-81 mo wheezing OR, 3.9 (95% CI, 2.0-7.9) for asthma at age 91 mo	No interaction identified between atopy and RSV infection ($P =$ 1.0 if outcome doctor- diagnosed asthma at age 91 mo; $P = 0.09$ if outcome wheezing at age 69-81 mo and $P =$ 0.6 if outcome persistent wheeze at 30-42 and 69-81 mo)

Jackson et al ^{E2} (2008) USA	COAST birth cohort N = 259 (6 y)	Nasal lavage, culture and PCR for multiple viruses Viral studies at 2, 4, 6, 9, and 12 mo and in illness	Annual blood specific IgE; SPT at age 5 y	Current asthma: Doctor- diagnosed asthma and treatment at age 6 y	Parental asthma, birth month, weight, dog and cat exposure, older siblings, passive smoke, daycare, breast-feeding	Age 1: In atopic individuals, OR for asthma with RV wheezing = 7.5 In nonatopic individuals, OR for asthma with RV wheezing = 2.49 Age 3: In atopic individuals, OR for asthma with RV wheezing = 23.1 In nonatopic individuals, OR for asthma with RV wheezing = 26.6	2008: Aeroallergen sensitization did not modify relationship between RV wheezing illness and asthma development ($P =$ 0.99 in year 3 and $P =$ 0.61 in year 1) 2011: No data on interactions
Rubner et al ^{E8} (2017) USA	N = 217 (13 y)					Age 6: OR for asthma = 45 (HRV + aeroallergen sensitization at <3 y) vs 7.8 for HRV, 3.9 for aeroallergen sensitization Age 8: OR for asthma = 20 (HRV + aeroallergen sensitization at <3 y) vs 8.1 for HRV, 2.9 for aeroallergen sensitization Age 11: OR for asthma = 14 (HRV + aeroallergen sensitization at <3 y) vs 6.2 for HRV, 2.9 for aeroallergen sensitization Age 13: OR for asthma = 14 (HRV + aeroallergen sensitization at <3 y) vs 3.0 for HRV, 2.7 for aeroallergen sensitization	Additive effects of early life aeroallergen sensitization and RV wheezing on asthma risk at adolescence
Kusel et al ^{E3} (2007) Australia	N = 198 Birth cohort High atopy risk Follow-up to age 5 y in 2007)	Viral symptoms NPA within 48 h with PCR virus identification URTI and LRTI on history Febrile if temperature = 38°C+	SPT at 6 mo, 2 y, 5 y (wheal of 3 mm+ = atopy at 5; 2 mm at 6 mo and 2 y)	History of wheeze and doctor-diagnosed asthma; data collected annually	Sex, breast-feeding, pets, older siblings, ETS, daycare	2007: at age 5 y: Nonatopic individuals: Any wheezy/febrile LRTI year 1: OR, 1.0 (95% CI, 0.3-3.4) for current wheeze at age 5 Atopic individuals: Atopy by age 2: OR = 3.9 (95% CI, 1.4-10.5) Atopic individuals after age 2: OR = 0.7 (95% CI, 0.1-3.9)	2007: Interaction between wheezy LRTI in year 1 and atopy by age 2 y:
Kusel et al ^{E9} (2012) Australia	*(Subset followed up at age 10 y in 2012; n = 147)	Viral symptoms NPA within 48 h with PCR virus identification URTI and LRTI on history Febrile if temperature = 38°C+ on 2 occasions	SPT at 10 y to panel of 7 food and aeroallergens (wheal of 3 mm+ = atopy)	History of wheeze in 1 y before 10-y visit; doctor-diagnosed asthma; current asthma = diagnosis + PW or medications in 1 y before review	Sex, breast-feeding, pets, older siblings, ETS, daycare	2012: At age 10 y: Atopy by 2 y and LRTI and fever: OR for asthma at age 10 y = 4.92 (95% CI 2.59-9.36) Atopy after 2 y: OR, 1.13 (95% CI, 0.19- 6.79) Wheezy or febrile LRTI and atopy by age 2 y: OR for asthma at age 10 y = 2.92; atopy after 2 y: OR = 0.61 (95% CI, 0.09-3.97)	2012: Greater effect for febrile LRTI Interaction between early and persistent atopy and febrile LRTI in year 1

(continued)

TABLE E1. (Continued)

Author, date Country	Population	Exposure: Infection	Allergy/Atopy	Outcome: Wheezing/ asthma	Confounders	Results	Author's conclusion
Lukkarinen et al ^{1E4} (2017) Finland	N = 127 7-y follow-up First severe wheezing episode Vinku and Vinku 2 studies Prospective cohort Median age at entry = 11 mo	Viral identification via nasopharyngeal aspirate PCR Culture, antigen detection, and serology	Allergen specific IgE, blood eosinophil counts Spirometry at age 8 y— pre- and postbronchodilator BHR in Vinku2	Asthma at age 8 y Current asthma with symptoms or treatment previous 12 mo	Passive smoke, breast- feeding, family history for asthma and atopy, sibling number	Asthma at age 8 y: Sensitization and HRV = 10 of 17 (59%) Sensitization or HRV = 12 of 56 (24%) No sensitization or HRV = 14 of 56 (25%)	No comment regarding interaction between HRV and sensitization

ALSPAC, Avon Longitudinal Study of Parents And Children; BHR, bronchial hyperresponsiveness; COPSAC, Copenhagen Prospective Studies of Asthma in Childhood; ETS, environmental tobacco smoke; HPA, hypopharyngeal aspirate; LRTI, lower respiratory tract infection; NPA, nasopharyngeal aspirate; PW, persistent wheeze; RV, rhinovirus; URTI, upper respiratory tract infection.

TABLE E2. Newcastle-Ottawa Quality Assessment Scale for included studies

Author (year) Study design	Oddy et al ^{E5} (2002) cohort*	Nafstad et al ^{E6} (2005) cohort*	Henderson et al ^{E7} (2005) cohort	Kusel et al ^{E3} (2007) cohort*	Kusel et al ^{E9} (2012) cohort	Bønnelykke et al ^{E1} (2015) cohort*	Jackson et al ^{E2} (2008) cohort	Rubner et al ^{E8} (2017) cohort	Lukkarinen et al ^{E4} (2017) cohort
Newcastle-Ottawa Scale points	7/9	5/9	6/9	8/9	8/9	7/9	8/9	8/9	8/9
Case definition	NA	NA	NA	NA	NA	NA	NA	NA	NA
Definition of controls	NA	NA	NA	NA	NA	NA	NA	NA	NA
Sample representativeness	1	1	1	1	1	1	1	1	1
Sample size	NA	NA	NA	NA	NA	NA	NA	NA	NA
Nonrespondents	NA	NA	NA	NA	NA	NA	NA	NA	NA
Ascertainment of exposure	1	0	1	1	1	0	1	1	1
Comparability	1	1	1	1	1	1	1	1	1
Assessment of outcome	0	0	0	0	1	1	1	1	1
Statistical test	NA	NA	NA	NA	NA	NA	NA	NA	NA
Selection of nonexposed cohort	1	1	1	1	1	1	1	1	1
Outcome not present at start of study	1	1	1	1	1	1	1	1	1
Adequate follow-up period	1	1	1	1	1	1	1	1	1
Adequacy of cohort follow-up	1	0	0	1	1	1	1	1	1

NA, (Not applicable) entered where the specific criterion is not included in the scale appropriate for the study design.

*Studies included in meta-analysis.

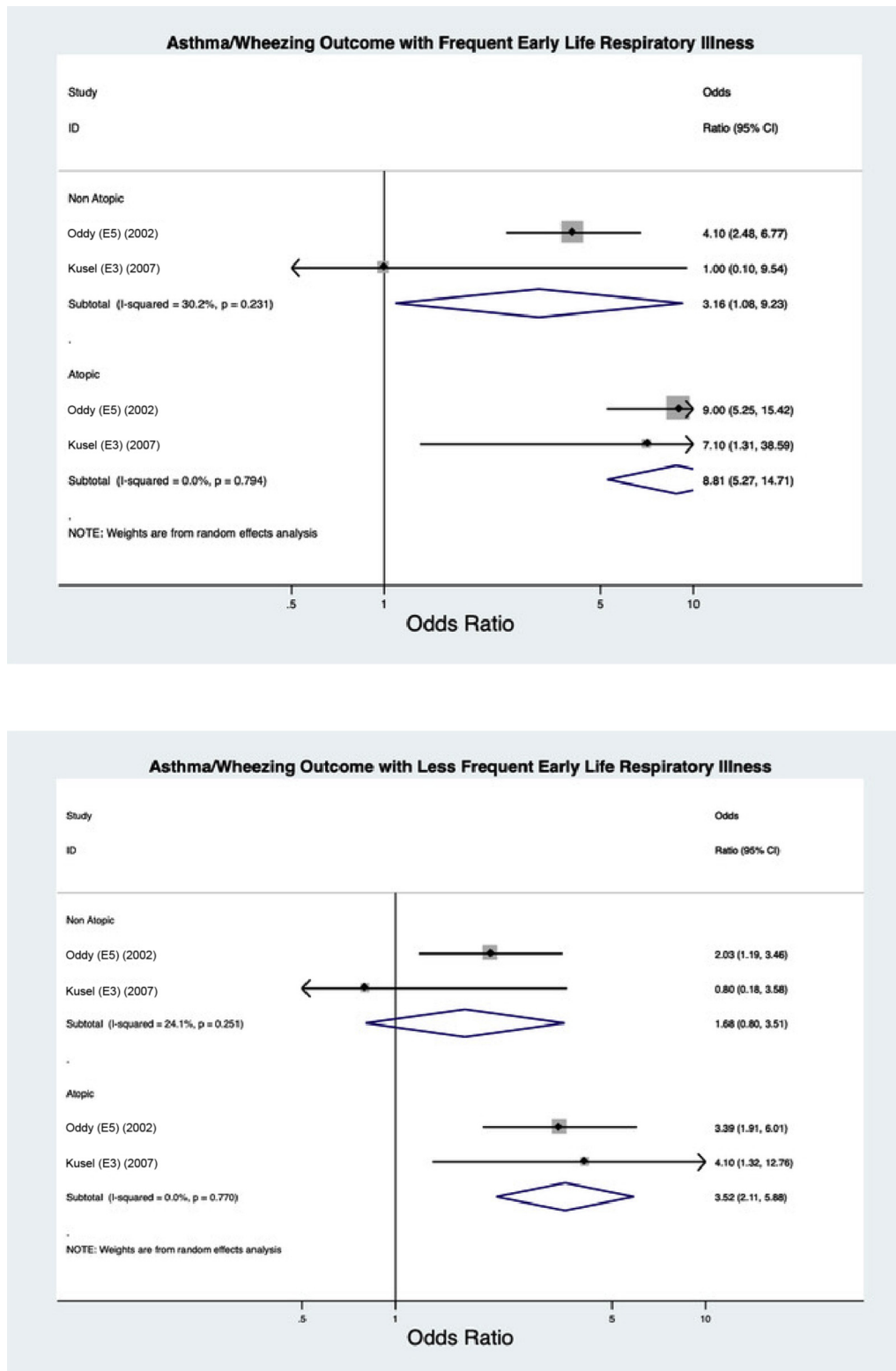


FIGURE E1. Meta-analysis of the association of early life respiratory illness with asthma/wheeze outcome across nonatopic and atopic subjects by frequency of viral respiratory illness.

REFERENCES

- E1. Bønnelykke K, Vissing NH, Sevelsted A, Johnston SL, Bisgaard H. Association between respiratory infections in early life and later asthma is independent of virus type. *J Allergy Clin Immunol* 2015;136:81-86.e4.
- E2. 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.
- E3. Kusel MM, de Klerk NH, Keadze 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.
- E4. Lukkariinen M, Koistinen A, Turunen R, Lehtinen P, Vuorinen T, Jartti T. Rhinovirus-induced first wheezing episode predicts atopic but not nonatopic asthma at school age. *J Allergy Clin Immunol* 2017;140:988-95.
- E5. Oddy WH, de Klerk NH, Sly PD, Holt PG. The effects of respiratory infections, atopy, and breastfeeding on childhood asthma. *Eur Respir J* 2002;19:899-905.
- E6. Nafstad P, Brunekreef B, Skrandal A, Nystad W. Early respiratory infections, asthma, and allergy: 10-year follow-up of the Oslo birth cohort. *Pediatrics* 2005;116:e255-62.
- E7. 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.
- E8. Rubner FJ, Jackson DJ, Evans MD, Gangnon RE, Tisler CJ, Pappas TE, et al. Early life rhinovirus wheezing, allergic sensitization, and asthma risk at adolescence. *J Allergy Clin Immunol* 2017;139:501-7.
- E9. Kusel MM, Keadze T, Johnston SL, Holt PG, Sly PD. Febrile respiratory illnesses in infancy and atopy are risk factors for persistent asthma and wheeze. *Eur Respir J* 2012;39:876-82.