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The relationship between RSV bronchiolitis and recurrent wheeze: The chicken *and* the egg

L. Bont^{a,*}, O. Ramilo^b^a University Medical Centre Utrecht, Department of Pediatrics, PO Box 85090, 3508 AB, Utrecht, Netherlands^b Division of Pediatric Infectious Diseases, Center for Vaccines and Immunity, Research Institute at Nationwide Children's Hospital, Ohio State University College of Medicine, 700 Children's Dr, W4022, Columbus, OH 43205, United States

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ABSTRACT

Respiratory syncytial virus bronchiolitis is the most frequent cause of infant hospitalization. RSV bronchiolitis is often followed by recurrent episodes of wheeze. Pathogenesis of RSV bronchiolitis as well as post-bronchiolitis wheeze are incompletely understood. The aim of this review is to provide a brief overview of our current understanding of the complex pathogenesis of RSV bronchiolitis and post-bronchiolitis wheeze. Two non-exclusive hypotheses exist, which are paraphrased for this review as “the chicken and the egg”. First, we reviewed the pre-existent genetic, pulmonary and immunological mechanisms of RSV bronchiolitis and post-bronchiolitis wheeze. Second, RSV as the causative virus of long-term airway morbidity is reviewed. Clearly, RSV infection is capable of causing direct damage to the airways and/or inducing long-term inappropriate immune responses to respiratory viruses or aero-allergens. It is concluded that intervention trials aimed at preventing RSV infections are required to establish the relative contribution of both RSV-induced and pre-existent mechanisms to the development of long-term airway disease following RSV bronchiolitis.

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1. Introduction

Respiratory syncytial virus bronchiolitis is the most common cause of hospitalization during infancy. In the general population 2% of all children are admitted for RSV bronchiolitis during the first year of life [1]. In preterm children with chronic lung disease, the risk of hospitalization for RSV infection may be as high as 33% [2]. Disease during RSV bronchiolitis is caused by complex mechanisms. Virus-induced cytopathology, virus-induced immunopathology and pre-existent susceptibility factors all contribute to symptoms observed during RSV bronchiolitis. RSV bronchiolitis is often followed by recurrent episodes of wheeze or post-bronchiolitis wheeze (PBW) [3]. The possible relationship between RSV and post-bronchiolitis wheeze is often described as a “chicken or egg” phenomenon. On one hand, RSV infection could be the cause of PBW by affecting airway structure and function or by inducing a long-term aberrant immune response to common respiratory viruses or aero-allergens. On the other hand,

RSV bronchiolitis may be the first indication of long-term airway morbidity in which host susceptibility factors determine the risk of both RSV bronchiolitis and recurrent wheeze. The aim of this review is to provide a brief overview of our current understanding of the complex pathogenesis of RSV bronchiolitis and PBW.

2. Epidemiologic studies

A number of retrospective studies since the 1970s have described a clear association between RSV bronchiolitis and increased risk for subsequent wheezing [4,5]. Because of the retrospective methodology of those initial studies, several authors raised questions about the significance of this association. However, in the last two decades a number of prospective studies conducted in different parts of the world and including children with different genetic backgrounds have confirmed a strong association between acute RSV bronchiolitis and PBW. Investigators in Arizona demonstrated that children with RSV bronchiolitis early in life had a significantly higher risk of wheezing until 6 years of age. It is important to mention that the majority of these children were not hospitalized during the acute episode of bronchiolitis, and yet developed subsequent PBW. Likewise, studies conducted in Sweden showed that children hospitalized for RSV bronchiolitis had

* Corresponding author.

E-mail address: L.bont@umcutrecht.nl (L. Bont).

increased risk of wheezing and asthma up until 18 years of age compared with age-matched controls [6–8]. In this particular cohort, in addition to PBW, patients had an increased risk for atopy, which has not been observed in other post-bronchiolitis study cohorts. More recently, studies conducted in Finland [9–11] also showed very similar and consistent results and further demonstrated the impact of PBW in quality of life [12].

3. Pre-existent mechanisms of RSV bronchiolitis and long-term wheeze

3.1. Genetic susceptibility

The Danish national birth cohort study has attempted to define to what extent the risk of hospitalization for RSV bronchiolitis during the first year of life is as explained by genetic variation [13]. In 12,346 homozygous and heterozygous twins, the concordance rate for hospitalization for RSV infection was 0.66 and 0.53, respectively. It was calculated that the heritability of RSV bronchiolitis is 16%. During the last decade genetic studies have evolved and demonstrated the existence of various susceptibility genes associated with bronchiolitis. Initially, one single nucleotide polymorphism (SNP) of one candidate gene, for example the IL-4-589T promoter SNP, was studied in relation to the risk of severe RSV bronchiolitis [14]. Although in many occasions, the functionality of these SNP was unknown. Later, complex genetic studies were performed in which several hundred SNPs were studied [15]. Forton et al. studied 113 single SNPs in 11 genes at 5q31 cytokine cluster demonstrating an association between the IL13 and the SLC22A5 gene and a risk of severe RSV infection. [16]. Recently, genetic mechanisms of disease were specifically studied in preterm infants showing specific associations between airway remodelling genes and risk of severe RSV infection in preterms [17]. These genetic studies have provided basic insight into RSV pathogenesis as recently reviewed [3,18]. Specific genes that have been associated with the risk of RSV bronchiolitis can arbitrarily be divided into the following categories: 1) airway mucosal responses, 2) innate immune responses, 3) adaptive immune responses, 4) chemotaxis and 5) allergic responses. Global genetic analyses suggested that variation in innate immunity genes has the biggest impact on the risk of severe RSV infection [15]. A major concern of the SNP studies related to RSV bronchiolitis is that most genetic associations have not yet been confirmed. This is a critical issue, because this is required before any genetic association can be considered true. Confirmation of an association with RSV bronchiolitis was achieved for a few polymorphisms, including the TLR-4 Asp299Gly or Thr399Ile polymorphisms [19–22], the IL4 rs2243250 SNP [14,23] and a specific IL4/IL13 haplotype [16,24]. Future studies should not only aim at discovering new genes associated with RSV pathogenesis, but also at confirming previously reported associations. Finally, large genetic cohorts are required to perform genome wide association studies. Genome wide association studies are unbiased and will lead to the next level of understanding of the complex genetic mechanisms underlying RSV bronchiolitis.

3.2. Pulmonary susceptibility

Pulmonary function abnormalities may underlie both the risk of RSV bronchiolitis and recurrent wheeze. The population at the highest risk of RSV bronchiolitis are severely preterm infants with chronic lung disease [2]. In these children, severity of lung function abnormalities is associated with risk of severe RSV bronchiolitis [25]. Late preterms are also born with abnormal lung function, which is not always clinically manifest [26]. Probably, risk of RSV bronchiolitis in late preterms is dictated by the infant's lung function. Another population at risk of severe RSV bronchiolitis is infants with cystic fibrosis, although the absolute risk of RSV bronchiolitis has not

been studied in this population [27]. RSV infection in this population may even be more relevant, because it might be associated with long-term deterioration of lung function. In vitro and in vivo studies show that RSV facilitates *Pseudomonas* infection of airway epithelium [28,29]. In healthy term infants, lung function at birth is associated with the risk of wheezing illness during pre-school years [30]. Airway compliance was recently shown to be associated with the risk of rhinovirus-associated wheezing illness [31]. It is hypothesized that abnormal lung function at birth predisposes not only to severe rhinovirus infection, but also severe RSV infection.

3.3. Cardiovascular susceptibility

It has long been known that congenital heart anomalies are associated with the risk of RSV bronchiolitis. The risk appears highest in children with increased pulmonary flow. Surgical intervention may correct the anatomy of the heart, although it is not known how long it takes before the risk of RSV bronchiolitis becomes normal in this population. It is conceivable that this may take several weeks or even months before the vascular functions pulmonary and systemic have normalized. Down Syndrome is an independent risk factor of RSV bronchiolitis [32]. Pulmonary hypertension is strongly associated with Down Syndrome, which suggests that abnormal pulmonary vascular function plays a role in the susceptibility of this specific high-risk population [33]. However, other mechanisms may also be relevant in the pathogenesis of RSV bronchiolitis in children with Down Syndrome, such as abnormal upper airway anatomy [34] or abnormal adaptive and innate immune responses [35].

3.4. Immunological susceptibility

Immature innate and adaptive immune responses have been suggested to play an important role in susceptibility to RSV bronchiolitis [36]. Children are born with immature production of Th1 cytokines, which is probably required to sustain intra-uterine life, but a risk factor for infection with intracellular pathogens during infancy [37]. It was shown that cord blood low Th1 interferon (IFN)- γ predisposes children to wheezing illness during the first years of life [38]. Antigen-presenting cell (APC) production of the Th1 directing cytokine IL-12 is also immature at birth [39], which further stresses the relative immature immune status of the neonate. A recent study specifically compared characteristics of cord blood transcripts in children who would later develop RSV bronchiolitis and those who would not [40]. Interestingly, it was shown that not only genes involved in innate and adaptive immune responses, but also genes regulating actin cytoskeleton formation were associated with risk of RSV bronchiolitis.

4. RSV as the cause of long-term airway morbidity

In addition to all the different factors (genetic, pulmonary, cardiac and immunologic) that are associated with increased susceptibility to develop severe RSV bronchiolitis, it is thought that the virus itself has unique characteristics that play a significant role in its ability to cause (or to be associated with) long-term pulmonary sequelae. Although the epidemiologic studies demonstrated a strong association between RSV bronchiolitis and PBW, those observational studies do not allow dissecting the many factors involved. To better define the role of the virus itself and to separate it from other factors, it is necessary to take other approaches.

The mouse model of RSV has been adapted to study the role of the virus in the long-term pulmonary sequelae. Mice are inoculated intranasally with RSV and using a non-invasive plethysmograph, they can be followed and their pulmonary function monitored for weeks after the acute infection. Using this approach there is clear evidence that RSV alone – and without any added factor – causes airway

hyperreactivity and chronic airway inflammation for months after the initial RSV infection [41]. These long-term pulmonary sequelae induced by RSV are independent of the mouse strain [42]. More recent studies in this model have demonstrated that airway hyperreactivity and chronic inflammation are associated with detection of persistent RSV RNA in the lung, thus suggesting a potential novel virus-dependent mechanism for PBW [43]. The establishment of a model of RSV-induced hyperreactivity and chronic airway inflammation provided experimental evidence of the link between RSV bronchiolitis and PBW and simultaneously allowed the implementation of therapeutic interventions to further assess the role of the virus itself. The administration of a single dose of anti-RSV neutralizing monoclonal antibodies (palivizumab or motavizumab) was associated with significant reductions in long-term airway hyperreactivity and pulmonary inflammation [42,44], indicating that therapeutic and/or preventing interventions directed against the virus in the acute phase can provide long term benefit. Indeed, a recent double cohort study conducted in Europe demonstrated that premature infants with no CLD who received prophylaxis with palivizumab had a significant reduction in wheezing episodes compared with control premature infants who did not receive prophylaxis [45].

This study further confirms the overall hypothesis that RSV – the virus itself – plays a significant role in the origin of PBW, and more importantly establishes the proof of principle that interventions directed to prevent or modify acute RSV bronchiolitis can provide a long-term benefit reducing recurrent wheezing episodes in childhood.

5. Conclusion

Numerous epidemiologic studies have demonstrated a strong association between RSV bronchiolitis early in life and the development of post-bronchiolitis wheeze (PBW). The mechanisms responsible for increased susceptibility to develop severe RSV bronchiolitis are complex and multifactorial. Studies have confirmed that different genetic, pulmonary, cardiac and immunological factors influence the risk for severe bronchiolitis and PBW. In addition, experimental studies showed that the virus itself plays a major role in the development of airway hyperactivity and chronic inflammation. Further support for the critical role of RSV itself in the development of PBW is derived from a recent parallel cohort study that demonstrated that premature children who received prophylaxis with an anti-RSV monoclonal antibody had significantly less frequent wheezing episodes than untreated age-matched controls.

Conflict of interest

Both authors received speaker's fees from Abbott.

References

- [1] Smyth RL, Openshaw PJ. Bronchiolitis. *Lancet* 2006;368(9532):312–22.
- [2] Broughton LS, Bhat R, Roberts A, Zuckerman M, Rafferty G, Greenough A. Diminished lung function, RSV infection and respiratory morbidity in prematurely born infants. *Arch Dis Child* 2006;91:26–30.
- [3] Tregoning JS, Schwarze J. Respiratory viral infections in infants: causes, clinical symptoms, virology, and immunology. *Clin Microbiol Rev* 2010;23(1):74–98.
- [4] Rooney JC, Williams HE. The relationship between proved viral bronchiolitis and subsequent wheezing. *J Pediatr* 1971;79(5):744–7.
- [5] Perez-Yarza EG, Moreno A, Lazaro P, Mejias A, Ramilo O. The association between respiratory syncytial virus infection and the development of childhood asthma: a systematic review of the literature. *Pediatr Infect Dis J* 2007;26(8):733–9.
- [6] Sigurs N, Bjarnason R, Sigurbergsson F, Kjellman B. Respiratory syncytial virus bronchiolitis in infancy is an important risk factor for asthma and allergy at age 7. *Am J Respir Crit Care Med* 2000;161(5):1501–7.
- [7] Sigurs N, Gustafsson PM, Bjarnason R, Lundberg F, Schmidt S, Sigurbergsson F. Severe respiratory syncytial virus bronchiolitis in infancy and asthma and allergy at age 13. *Am J Respir Crit Care Med* 2005;171(2):137–41.
- [8] Sigurs N, Aljassim F, Kjellman B, Robinson PD, Sigurbergsson F, Bjarnason R. Asthma and allergy patterns over 18 years after severe RSV bronchiolitis in the first year of life. *Thorax* 2010;65:1045–52.
- [9] Korppi M, Piippo-Savolainen E, Korhonen K, Remes S. Respiratory morbidity 20 years after RSV infection in infancy. *Pediatr Pulmonol* 2004;38(2):155–60.
- [10] Henderson J, Hilliard TN, Sherriff A, Stalker D, Al SN, 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(5):386–92.
- [11] Bont L, van Aalderen WM, Versteegh J, Brus F, Draaisma JT, Pekelharing-Berghuis M. Airflow limitation during respiratory syncytial virus lower respiratory tract infection predicts recurrent wheezing. *Pediatr Infect Dis J* 2001;20(3):277–82.
- [12] Bont L, Steijn M, van Aalderen WM, Kimpfen JL. Impact of wheezing after respiratory syncytial virus infection on health-related quality of life. *Pediatr Infect Dis J* 2004;23(5):414–7.
- [13] Thomsen SF, Stensballe LG, Skytthe A, Kyvik KO, Backer V, Bisgaard H. Increased concordance of severe respiratory syncytial virus infection in identical twins. *Pediatrics* 2008;121(3):493–6.
- [14] Choi EH, Lee HJ, Yoo T, Chanock SJ. A common haplotype of interleukin-4 gene IL4 is associated with severe respiratory syncytial virus disease in Korean children. *J Infect Dis* 2002;186(9):1207–11.
- [15] Janssen R, Bont L, Siezen CL, Hodemaekers HM, Ermers MJ, Doornbos G. Genetic susceptibility to respiratory syncytial virus bronchiolitis is predominantly associated with innate immune genes. *J Infect Dis* 2007;196(6):826–34.
- [16] Forton JT, Rowlands K, Rockett K, Hanchard N, Herbert M, Kwiatkowski DP. Genetic association study for RSV bronchiolitis in infancy at the 5q31 cytokine cluster. *Thorax* 2009;64(4):345–52.
- [17] Siezen CL, Bont L, Hodemaekers HM, Ermers MJ, Doornbos G, Van't Slot R. Genetic susceptibility to respiratory syncytial virus bronchiolitis in preterm children is associated with airway remodeling genes and innate immune genes. *Pediatr Infect Dis J* 2009;28(4):333–5.
- [18] Miyairi I, DeVincenzo JP. Human genetic factors and respiratory syncytial virus disease severity. *Clin Microbiol Rev* 2008;21(4):686–703.
- [19] Tal G, Mandelberg A, Dalal I, Cesar K, Somekh E, Tal A. Association between common Toll-like receptor 4 mutations and severe respiratory syncytial virus disease. *J Infect Dis* 2004;189(11):2057–63.
- [20] Puthothu B, Forster J, Heinzmann A, Krueger M. TLR-4 and CD14 polymorphisms in respiratory syncytial virus associated disease. *Dis Markers* 2006;22(5–6):303–8.
- [21] Awomoyi AA, Rallabhandi P, Pollin TI, Lorenz E, Szein MB, Boukhvalova MS. Association of TLR4 polymorphisms with symptomatic respiratory syncytial virus infection in high-risk infants and young children. *J Immunol* 2007;179(5):3171–7.
- [22] Lofgren J, Marttila R, Renko M, Ramet M, Hallman M. Toll-like receptor 4 Asp299Gly polymorphism in respiratory syncytial virus epidemics. *Pediatr Pulmonol* 2010;45(7):687–92.
- [23] Hoebee B, Rietveld E, Bont L, Oosten M, Hodemaekers HM, Nagelkerke NJ. Association of severe respiratory syncytial virus bronchiolitis with interleukin-4 and interleukin-4 receptor alpha polymorphisms. *J Infect Dis* 2003;187(1):2–11.
- [24] Puthothu B, Krueger M, Forster J, Heinzmann A. Association between severe respiratory syncytial virus infection and IL13/IL4 haplotypes. *J Infect Dis* 2006;193(3):438–41.
- [25] Broughton S, Bhat R, Roberts A, Zuckerman M, Rafferty G, Greenough A. Diminished lung function, RSV infection, and respiratory morbidity in prematurely born infants. *Arch Dis Child* 2006;91(1):26–30.
- [26] Friedrich L, Pitrez PM, Stein RT, Goldani M, Tepper R, Jones MH. Growth rate of lung function in healthy preterm infants. *Am J Respir Crit Care Med* 2007;176(12):1269–73.
- [27] Fauroux B. Special populations. *Paediatr Respir Rev* 2009;10(Suppl 1):21–2.
- [28] van Ewijk BE, Wolfs TF, Aerts PC, Van Kessel KP, Fleer A, Kimpfen JL. RSV mediates *Pseudomonas aeruginosa* binding to cystic fibrosis and normal epithelial cells. *Pediatr Res* 2007;61(4):398–403.
- [29] de Vrankrijker AM, Wolfs TF, Ciofu O, Høiby N, van der Ent CK, Poulsen SS. Respiratory syncytial virus infection facilitates acute colonization of *Pseudomonas aeruginosa* in mice. *J Med Virol* 2009;81(12):2096–103.
- [30] Martinez FD, Morgan WJ, Wright AL, Holberg CJ, Taussig LM. Diminished lung function as a predisposing factor for wheezing respiratory illness in infants. *N Engl J Med* 1988;319:1112–7.
- [31] van der Zalm MM, Uiterwaal CS, Wilbrink B, Koopman M, Verheij TJ, Van der Ent CK. The influence of neonatal lung function on rhinovirus associated wheeze. *Am J Respir Crit Care Med* 2010;183:262–7.
- [32] Bloemers BL, van Furth AM, Weijerman ME, Gemke RJ, Broers CJ, Kimpfen JL. Down syndrome: a novel risk factor for respiratory syncytial virus bronchiolitis—a prospective birth-cohort study. *Pediatrics* 2007;120(4):e1076–81.
- [33] Shah PS, Hellmann J, Adatia I. Clinical characteristics and follow up of Down syndrome infants without congenital heart disease who presented with persistent pulmonary hypertension of newborn. *J Perinat Med* 2004;32(2):168–70.
- [34] Uong EC, McDonough JM, Tayag-Kier CE, Zhao H, Haselgrove J. Magnetic resonance imaging of the upper airway in children with Down syndrome. *Am J Respir Crit Care Med* 2001;163(3 Pt 1):731–6.
- [35] Bloemers BL, Broers CJ, Bont L, Weijerman ME, Gemke RJ, van Furth AM. Increased risk of respiratory tract infections in children with Down syndrome: the consequence of an altered immune system. *Microbes Infect* 2010;12(11):799–808.
- [36] Bont L, Kimpfen JL. Immunological mechanisms of severe respiratory syncytial virus bronchiolitis. *Intensive Care Med* 2002;28(5):616–21.
- [37] Wilson CB. Immunological basis for increased susceptibility of the neonate to infection. *J Pediatr* 1986;108:1–12.

- [38] Copenhaver CC, Gern JE, Li Z, Shult PA, Rosenthal LA, Mikus LD. Cytokine response patterns, exposure to viruses, and respiratory infections in the first year of life. *Am J Respir Crit Care Med* 2004;170(2):175–80.
- [39] Belderbos ME, van Bleek GM, Levy O, Blanken MO, Houben ML, Schuijff L. Skewed pattern of Toll-like receptor 4-mediated cytokine production in human neonatal blood: low LPS-induced IL-12p70 and high IL-10 persist throughout the first month of life. *Clin Immunol* 2009;133(2):228–37.
- [40] Fjaerli HO, Bukholm G, Skjaeret C, Holden M, Nakstad B. Cord blood gene expression in infants hospitalized with respiratory syncytial virus bronchiolitis. *J Infect Dis* 2007;196(3):394–404.
- [41] Jafri HS, Chavez-Bueno S, Mejias A, Gomez AM, Rios AM, Nassi SS. Respiratory syncytial virus induces pneumonia, cytokine response, airway obstruction, and chronic inflammatory infiltrates associated with long-term airway hyperresponsiveness in mice. *J Infect Dis* 2004;189(10):1856–65.
- [42] Mejias A, Chavez-Bueno S, Rios AM, Aten MF, Raynor B, Peromingo E. Comparative effects of two neutralizing anti-respiratory syncytial virus (RSV) monoclonal antibodies in the RSV murine model: time versus potency. *Antimicrob Agents Chemother* 2005;49(11):4700–7.
- [43] Estripeaut D, Torres JP, Somers CS, Tagliabue C, Khokhar S, Bhoj VG. Respiratory syncytial virus persistence in the lungs correlates with airway hyperreactivity in the mouse model. *J Infect Dis* 2008;198(10):1435–43.
- [44] Mejias A, Chavez-Bueno S, Rios AM, Saavedra-Lozano J, Fonseca Aten M, Hatfield J. Anti-respiratory syncytial virus (RSV) neutralizing antibody decreases lung inflammation, airway obstruction, and airway hyperresponsiveness in a murine RSV model. *Antimicrob Agents Chemother* 2004;48(5):1811–22.
- [45] Simoes EA, Groothuis JR, Carbonell-Estrany X, Rieger CH, Mitchell I, Fredrick LM. Palivizumab prophylaxis, respiratory syncytial virus, and subsequent recurrent wheezing. *J Pediatr* 2007;151(1):34–42.