BRIEF REPORT

Smoke Exposure, Cytokine Levels, and Asthma Visits in Children Hospitalized for Bronchiolitis

Amy L. Willis, MD,[a] Angela Moss, MS,[a,b] Michelle Torok, PhD,[a,b] Michelle Lowary, MS,[c] Jonathan D. Klein, MD, MPH,[d,e] Karen M. Wilson, MD, MPH[e,f]

ABSTRACT

BACKGROUND AND OBJECTIVE: To determine if cigarette smoke exposure, marijuana smoke exposure, or cytokine levels at admission to the hospital for bronchiolitis are associated with follow-up visits for asthma.

METHODS: We enrolled a cohort of children aged 31 days to 2 years who were hospitalized with bronchiolitis from January 2013 to April 2014. Data included the results of a baseline survey about children's health and demographics, nasal wash samples, the results of a 6-month postdischarge follow-up survey, and a chart review. Nasal wash samples were tested for interleukin (IL)-6, IL-13, and tumor necrosis factor α (TNF-α); values were categorized for analysis. χ^2, Fisher's exact, and Wilcoxon rank tests were done to test bivariable differences; all analyses were done using SAS.

RESULTS: We approached 180 families for enrollment; 99 consented to participate, and 74% of these completed follow-up surveys. Half of those with high levels of IL-13 had follow-up visits for asthma, whereas only 4.2% of those with low levels had follow-up visits for asthma ($P = .02$). Marijuana exposure was reported for 12.5% ($n = 7$) of study participants. There was a significant association between marijuana exposure and TNF-α levels ($P = .03$).

CONCLUSIONS: Our study revealed an association between IL-13 and follow-up visits for asthma in children who were hospitalized with bronchiolitis. We found an association between family-reported marijuana smoke exposure and detectable but lower levels of TNF-α. Further research is needed to study these relationships.

[a]Department of Pediatric Hospital Medicine and [b]Adult and Child Center for Outcomes Research and Delivery Science, School of Medicine, University of Colorado, Aurora, Colorado; [c]Children's Hospital Colorado, Aurora, Colorado; [d]Department of Pediatrics, University of Illinois at Chicago, Chicago, Illinois; [e]American Academy of Pediatrics Julius B Richmond Center of Excellence, Elk Grove Village, Illinois; and [f]Department of Pediatrics, Icahn School of Medicine at Mount Sinai, New York, New York

www.hospitalpediatrics.org
DOI:https://doi.org/10.1542/hpeds.2018-0067

Address correspondence to Amy L. Willis, MD, Department of Pediatric Hospital Medicine, School of Medicine, University of Colorado, 13123 E. 16th Ave, B302, Aurora, CO 80045. E-mail: amy.willis@childrenscolorado.org

HOSPITAL PEDIATRICS (ISSN Numbers: Print, 2154-1663; Online, 2154-1671).

FINANCIAL DISCLOSURE: The authors have indicated they have no financial relationships relevant to this article to disclose.

FUNDING: Funded by the Flight Attendant Medical Research Institute through a grant to the American Academy of Pediatrics Julius B. Richmond Center of Excellence and the Children's Hospital Colorado Research Institute.

POTENTIAL CONFLICT OF INTEREST: The authors have indicated they have no potential conflicts of interest to disclose.

Dr Willis conducted the chart review of patients, drafted the initial manuscript, and revised the manuscript; Ms Moss and Dr Torok performed initial and statistical analyses and reviewed and revised the manuscript; Ms Lowary coordinated and supervised data collection and reviewed and revised the manuscript; Dr Klein reviewed and revised the manuscript; Dr Wilson conceptualized and designed the study and reviewed and revised the manuscript; and all authors approved the final manuscript as submitted.

Respiratory illness is the most common reason for pediatric hospital admissions in the United States. T helper cell (Th) 2 cytokine predominance, which is associated with atopy, is associated with viral lower respiratory tract infections.[1] Many cytokines are implicated in the immune response to respiratory viruses, including tumor necrosis factor α (TNF-α), interferon-γ (IFN-γ), interleukin (IL)-4, and IL-6.[2–4]

Many children with severe bronchiolitis go on to develop asthma.[5] Asthma has been associated with both Th1 and Th2 inflammation. Studies reveal that high levels of Th2 cytokines are related to asthma symptoms in children.[6–8] Researchers are conflicted on whether children with respiratory viral illnesses have a Th2 dominant cytokine response[1,9,10] and the relationship between Th2 cytokines in bronchiolitis and the development of asthma.[11–13] Tobacco smoke exposure increases the risk of developing asthma.[14] Additionally, there is evidence that secondhand smoke exposure in children is related to differing levels of inflammatory cytokines.[15]

In this study, we examined the association between cytokine levels in children with bronchiolitis and an asthma visit 6 months after discharge. We hypothesized that children with elevated Th2 cytokines (IL-4, IL-6 and IL-13) would be more likely to have a follow-up asthma visit compared with those with normal levels and that tobacco and marijuana smoke exposure would be associated with cytokine levels and asthma follow-up visits.

METHODS

We enrolled children aged 31 days to 2 years who were hospitalized at a free-standing children's hospital with a diagnosis of bronchiolitis between January 2013 and April 2014. After giving consent, parents completed a survey about their children's health, family demographics, and children's exposure to tobacco smoke; marijuana exposure was added after it was legalized in the state. Nasal wash samples were collected from each child. Six months after discharge, parents completed follow-up surveys, and chart reviews were completed by a trained abstractor; a report of a follow-up visit for asthma was recorded. Nasal wash samples were tested for IL-6, IL-13, TNF-α, and IFN-γ by using enzyme-linked immunosorbent assay. This study was approved by the Colorado Multiple Institutional Review Board.

TABLE 1 Characteristics of Children With Bronchiolitis Hospitalized at Children's Hospital Colorado (January 2013–April 2014)

Variables	Overall (N = 97)	Low IL-13 (N = 91)	High IL-13 (N = 6)	P
Age at admission, y				.10
Median (IQR)	0.6 (0.3–1.2)	0.6 (0.3–1.2)	1.2 (0.7–1.4)	
Sex, *n* (%)				.06
Girls	34 (35.1)	34 (37.4)	0 (0.0)	
Boys	63 (64.9)	57 (62.6)	6 (100.0)	
Race, *n* (%)				.69
White	79 (81.4)	73 (80.2)	6 (100.0)	
African American	4 (4.1)	4 (4.4)	0 (0.0)	
Other	3 (3.1)	3 (3.3)	0 (0.0)	
Multiracial	11 (11.3)	11 (12.1)	0 (0.0)	
Ethnicity, *n* (%)				.98
Non-Hispanic and/or non-Latino	49 (50.5)	46 (50.5)	3 (50.0)	
Hispanic and/or Latino	48 (49.5)	45 (49.5)	3 (50.0)	

IQR, interquartile range.

TABLE 2 Characteristics of Children With Bronchiolitis Hospitalized at Children's Hospital Colorado (January 2013–April 2014) by Level of IL-13

Variables	Overall (N = 75)	Low IL-13 (N = 71)	High IL-13 (N = 4)	P
	n (%)	*n* (%)	*n* (%)	
Self-reported exposure to cigarette smoke in last 24 h				.58
No	70 (93.3)	66 (93.0)	4 (100.0)	
Yes	5 (6.7)	5 (7.0)	0 (0.0)	
Follow-up visit for asthma or reactive airway disease				.05
No	67 (89.3)	65 (91.5)	2 (50.0)	
Yes	8 (10.7)	6 (8.5)	2 (50.0)	
Follow-up visit for asthma				.02
No	70 (93.3)	68 (95.8)	2 (50.0)	
Yes	5 (6.7)	3 (4.2)	2 (50.0)	
Oral steroids prescribed by provider in follow-up visit				.41
No	66 (88.0)	63 (88.7)	3 (75.0)	
Yes	9 (12.0)	8 (11.3)	1 (25.0)	
Inhaled steroids prescribed by provider in follow-up visit				.15
No	72 (96.0)	69 (97.2)	3 (75.0)	
Yes	3 (4.0)	2 (2.8)	1 (25.0)	
Albuterol prescribed by provider in follow-up visit				.21
No	57 (76.0)	55 (77.5)	2 (50.0)	
Yes	18 (24.0)	16 (22.5)	2 (50.0)	

Proportions and frequencies were used to describe categorical data; medians and interquartile ranges were used to describe continuous variables. χ^2 or Fisher's exact tests were used for categorical variables and Wilcoxon rank tests were used for continuous variables to compare cytokine with cigarette smoke and marijuana exposures on baseline demographics and respiratory outcomes. Analyses were done by using SAS 9.4 (SAS Institute, Inc, Cary, NC).[16]

RESULTS

We approached 180 families for enrollment; 99 (55%) agreed to participate, and 74% of these completed the follow-up survey at 6 months. Nasal wash samples were obtained from 98 children (99%). Fifty-six families were asked about exposure to marijuana. Table 1 includes the baseline demographics of the participants. IL-6 and TNF-α, which had 24% ($n = 24$) and 34% ($n = 34$) of data values below the limit of detection (LOD), respectively, were categorized as undetectable ($<$ LOD), low (25th percentile–75th percentile for IL-6; 35th percentile–75th percentile for TNF-α), and high ($>$75th percentile). IL-13 and IFN-γ, which had 93% ($n = 91$) and 88% ($n = 87$) below the LOD, respectively, were categorized as low ($<$ LOD) and high ($\geq$ LOD).

Fifty percent of those with elevated IL-13 ($n = 4$) had a follow-up appointment for asthma compared with 4.2% ($n = 3$) of those with low levels ($P = .02$; Table 2). There were no differences in cytokine levels or asthma visits by tobacco smoke exposure (Table 3).

Of the 56 subjects who were asked about marijuana exposure, 12.5% ($n = 7$) reported exposure to marijuana (Table 3). Patients who were exposed to marijuana were more likely to have low detectable levels of TNF-α than those who were not exposed to marijuana (86% [$n = 6$] vs 35% [$n = 17$]; $P = .03$). There was no statistically significant relationship between marijuana smoke exposure and visits for asthma.

DISCUSSION

Children who were hospitalized for bronchiolitis with elevated IL-13 at

TABLE 3 Characteristics of Children With Bronchiolitis Hospitalized at Children's Hospital Colorado (January 2013–April 2014) by Parent-Reported Marijuana and Cigarette Exposure

Variables	Cigarette Data Cohort ($N = 98$)			Marijuana Data Cohort ($N = 56$)		
	No Cigarette Exposure ($n = 93$)	Cigarette Exposure ($n = 5$)	*P*	No Marijuana Exposure ($n = 49$)	Marijuana Exposure ($n = 7$)	*P*
	n (%)	*n* (%)		*n* (%)	*n* (%)	
IL-6 cytokine						.07
Nondetectable	23 (24.7)	1 (20.0)	.75	15 (30.6)	0 (0.0)	
Low	47 (50.5)	2 (40.0)	—	20 (40.8)	6 (85.7)	
High	23 (24.7)	2 (40.0)	—	14 (28.6)	1 (14.3)	
TNF-α cytokine						.03
Nondetectable	32 (34.4)	2 (40.0)	.61	19 (38.8)	0 (0.0)	
Low	38 (40.9)	1 (20.0)	—	17 (34.7)	6 (85.7)	
High	23 (24.7)	2 (40.0)	—	13 (26.5)	1 (14.3)	
IL-13 cytokine						.26
Nondetectable	86 (93.5)	5 (100.0)	.56	47 (95.9)	6 (85.7)	
High	6 (6.5)	0 (0.0)	—	2 (4.1)	1 (14.3)	
IFN-γ cytokine						.29
Nondetectable	82 (88.2)	5 (100.0)	.41	42 (85.7)	7 (100.0)	
High	11 (11.8)	0 (0.0)	—	7 (14.3)	0 (0.0)	
Follow-up visit for asthma or reactive airway disease						.49
No	66 (91.7)	2 (50.0)	.05	33 (89.2)	4 (100.0)	
Yes	6 (8.3)	2 (50.0)	—	4 (10.8)	0 (0.0)	
Parental report of wheezing more than twice in 1 wk in follow-up						.98
No	62 (86.1)	2 (50.0)	.12	28 (75.7)	3 (75.0)	
Yes	10 (13.9)	2 (50.0)	—	9 (24.3)	1 (25.0)	
Albuterol prescribed by provider in follow-up visit						.23
No	56 (77.8)	2 (50.0)	.20	27 (73.0)	4 (100.0)	
Yes	16 (22.2)	2 (50.0)	—	10 (27.0)	0 (0.0)	

admission were more likely to have a visit for asthma, suggesting that IL-13 levels at admission might be used to predict which children were most likely to develop asthma. The patients with higher IL-13 levels were also older, which may have affected the diagnosis of asthma. If researchers in future studies confirm the association with higher IL-13 and later asthma development, hospitalization for bronchiolitis could represent an opportunity to identify children who are at high risk. Future researchers should confirm the relationship between cytokines, smoke exposure, and asthma. Then, interventions that are used to prevent asthma from developing (such as medications)[17] or aggressive interventions for environmental contributors (such as tobacco smoke) could be investigated.

There was not a significant association between reported tobacco smoke exposure and cytokine levels or asthma visits; this may be because smoke exposure was reported by families, and self-reported exposure underestimates actual smoke exposure as measured by cotinine in biologic samples.[18]

We found that in children who had detectable levels of TNF-α, reported marijuana smoke exposure was associated with lower rather than higher TNF-α levels. Although the relationship is complicated, this may support studies revealing that tetrahydrocannabinol suppresses TNF-α maturation and secretion in a murine model.[19] Further research is needed to understand the impact of marijuana smoke exposure on children.

We had a small sample size with limited statistical power; we were unable to control for confounding variables. We only included children who were hospitalized with bronchiolitis; thus, our cohort was likely more severely ill and not representative of the general population. Cytokine levels may vary depending on the severity of illness, virus, fluid sampled, and laboratory technique; these were not controlled for. We did not have biologic measures of either tobacco or marijuana smoke exposure and thus likely misclassified some children as unexposed. Using the 6-month follow-up, we may not have identified all children who will get asthma, and an asthma diagnosis at this age is subjective. It is unclear if using cytokines in clinical care is feasible or practical.

CONCLUSIONS

Our study revealed an association between IL-13 and later follow-up visits for asthma in children who are hospitalized with bronchiolitis, which could be used to identify children who are at high risk. Further research is needed to understand this relationship and determine if identifying children who are at risk could lead to interventions to prevent asthma.

REFERENCES

1. Byeon JH, Lee JC, Choi IS, Yoo Y, Park SH, Choung JT. Comparison of cytokine responses in nasopharyngeal aspirates from children with viral lower respiratory tract infections. *Acta Paediatr*. 2009;98(4):725–730
2. Matsuda K, Tsutsumi H, Okamoto Y, Chiba C. Development of interleukin 6 and tumor necrosis factor alpha activity in nasopharyngeal secretions of infants and children during infection with respiratory syncytial virus. *Clin Diagn Lab Immunol*. 1995; 2(3):322–324
3. Gentile DA, Doyle WJ, Zeevi A, et al. Cytokine gene polymorphisms moderate illness severity in infants with respiratory syncytial virus infection. *Hum Immunol*. 2003;64(3):338–344
4. Murai H, Terada A, Mizuno M, et al. IL-10 and RANTES are elevated in nasopharyngeal secretions of children with respiratory syncytial virus infection. *Allergol Int*. 2007;56(2):157–163
5. Balekian DS, Linnemann RW, Hasegawa K, Thadhani R, Camargo CA Jr. Cohort study of severe bronchiolitis during infancy and risk of asthma by age 5 years. *J Allergy Clin Immunol Pract*. 2017;5(1):92–96
6. Quah PL, Huang CH, Shek LP, Chua KY, Lee BW, Kuo IC. Hyper-responsive T-cell cytokine profile in association with development of early childhood wheeze but not eczema at 2 years. *Asian Pac J Allergy Immunol*. 2014;32(1):84–92
7. Chkhaidze I, Zirakishvili D, Shavshvishvili N, Barnabishvili N. Prognostic value of TH1/TH2 cytokines in infants with wheezing in a three year follow-up study. *Pneumonol Alergol Pol*. 2016;84(3): 144–150
8. Vizmanos-Lamotte G, Moreno-Galdó A, Muñoz X, Gómez-Ollés S, Gartner S, Cruz MJ. Induced sputum cell count and cytokine profile in atopic and non-atopic children with asthma. *Pediatr Pulmonol*. 2013;48(11):1062–1069
9. Legg JP, Hussain IR, Warner JA, Johnston SL, Warner JO. Type 1 and type 2 cytokine imbalance in acute respiratory syncytial virus bronchiolitis. *Am J Respir Crit Care Med*. 2003;168(6):633–639
10. Chung JY, Han TH, Kim JS, Kim SW, Park CG, Hwang ES. Th1 and Th2 cytokine levels in nasopharyngeal aspirates from children with human bocavirus bronchiolitis. *J Clin Virol*. 2008;43(2): 223–225
11. Castro M, Schweiger T, Yin-DeClue H, et al. Cytokine response after severe respiratory syncytial virus bronchiolitis in early life. *J Allergy Clin Immunol*. 2008; 122(4):726–733.e3
12. Wright AL. Epidemiology of asthma and recurrent wheeze in childhood. *Clin Rev Allergy Immunol*. 2002;22(1):33–44
13. Melendi GA, Laham FR, Monsalvo AC, et al. Cytokine profiles in the respiratory tract during primary infection with human metapneumovirus, respiratory syncytial virus, or influenza virus in infants. *Pediatrics*. 2007;120(2). Available at: www.pediatrics.org/cgi/content/full/120/2/e410
14. Feleszko W, Zawadzka-Krajewska A, Matysiak K, et al. Parental tobacco smoking is associated with augmented IL-13 secretion in children with allergic asthma. *J Allergy Clin Immunol*. 2006; 117(1):97–102
15. Wilson KM, Wesgate SC, Pier J, et al. Secondhand smoke exposure and serum cytokine levels in healthy children. *Cytokine*. 2012;60(1):34–37

16. *SAS 9.4 [computer program]*. Cary, NC: SAS Institute; 2013
17. Bagnasco D, Ferrando M, Varricchi G, Passalacqua G, Canonica GW. A critical evaluation of anti-IL-13 and anti-IL-4 strategies in severe asthma. *Int Arch Allergy Immunol*. 2016;170(2):122–131
18. Connor Gorber S, Schofield-Hurwitz S, Hardt J, Levasseur G, Tremblay M. The accuracy of self-reported smoking: a systematic review of the relationship between self-reported and cotinine-assessed smoking status. *Nicotine Tob Res*. 2009;11(1):12–24
19. Zheng ZM, Specter S, Friedman H. Inhibition by delta-9-tetrahydrocannabinol of tumor necrosis factor alpha production by mouse and human macrophages. *Int J Immunopharmacol*. 1992;14(8):1445–1452