

Hospitalization for Respiratory Syncytial Virus Bronchiolitis and Disease Severity in Twins

Miri Dotan^{1,4}, Liat Ashkenazi-Hoffnung MD^{2,4}, Zmira Samra PhD^{3,4}, Gilat Livni MD^{1,4}, Havatzelet Yarden-Bilavsky MD^{1,4}, Jacob Amir MD^{2,4} and Efraim Bilavsky MD^{2,4}

Departments of Pediatrics ¹A and ²C, Schneider Children's Medical Center of Israel, and ³Department of Microbiology, Rabin Medical Center (Beilinson Campus), Petah Tikva, Israel
⁴Sackler Faculty of Medicine, Tel Aviv University, Ramat Aviv, Israel

ABSTRACT: **Background:** Respiratory syncytial virus (RSV) is a common cause of lower respiratory tract disease and hospitalization in infants and young children. Infants of multiple births, who are often premature, might be more susceptible to developing a more severe RSV infection than singletons.

Objective: To assess the impact of multiple births on the severity of RSV infection and define risk factors for acquiring RSV infection in infants of multiple birth.

Methods: Clinical data on infants hospitalized with RSV infection between 2008 and 2010 were retrospectively collected.

Results: Twins comprised 7.6% (66/875) of hospitalized infants with RSV bronchiolitis during the study period. Infants in the twin group were younger (122.4 ± 131.7 vs. 204.5 ± 278.8 days, $P = 0.014$), had a lower mean gestational age (35.3 ± 2.6 vs. 38.6 ± 2.5 weeks, $P < 0.001$), and were more likely to have been born prematurely compared with singleton infants (65.6% vs. 13%, $P < 0.001$). On a multivariable logistic regression analysis, young age, early gestational age and male gender were the only variables identified as risk factors for pediatric intensive care unit admission ($P < 0.001$, $P < 0.001$ and $P = 0.03$, respectively). In contrast, the mere fact of a child being a twin was not found to be a significant risk factor for disease severity. In addition, if one twin is hospitalized due to RSV infection, the other has a 34% chance of also being hospitalized with bronchiolitis. Young age was a significant risk factor for hospitalization of the second twin ($P < 0.001$).

Conclusions: Our findings suggest that twins hospitalized with RSV bronchiolitis do not have an increased risk for severe infection as compared to singletons. However, a twin of an infant hospitalized with RSV infection has a considerable risk of also being hospitalized with bronchiolitis, thus close monitoring is recommended.

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KEY WORDS: respiratory syncytial virus (RSV), bronchiolitis, twins, multiple births

factors for hospitalization or severe disease [2]. Infants of multiple births are frequently premature and thus might be more susceptible to acquiring RSV infection [3]. Moreover, infants of multiple births share common genetic as well as environmental factors, which play a role in the risk for severe disease [2].

However, while in some studies multiple birth was found to be a strong risk factor for hospitalization due to severe RSV infection [3,4], in other studies multiple birth was not associated with a higher rate of RSV-related hospitalizations [5,6]. Moreover, because studies concerning this specific subject in Israel are lacking, the question regarding the association of multiple birth and severe RSV infection in Israel remains unanswered.

The aims of the present study were to compare multiple-birth infants and singletons hospitalized for RSV infection, determine whether multiple-birth infants were at increased risk for severe illness, and further define risk factors for developing RSV infection in multiple-birth infants.

PATIENTS AND METHODS

The study was conducted at Schneider Children's Medical Center, a tertiary university-affiliated pediatric hospital. A retrospective design was used. The study group comprised all children hospitalized between 1 January 2008 and 31 December 2010 with a positive RSV antigen as tested by enzyme-linked immunoassay in nasopharyngeal aspirates. Chronic disease was defined as the presence of pulmonary or cardiac dysfunction requiring medical therapy, or congenital/acquired immune deficiency. There were no exclusion criteria. For the purpose of the study, severe infection was defined as one that required hospitalization in the pediatric intensive care unit.

The following data were obtained: gestational history including multiple or singleton birth, patient age and gender, birth weight and gestational age, history of chronic disease, treatment received during hospitalization, and laboratory results. The clinical and laboratory findings were compared between multiple-birth infants and singletons.

RSV = respiratory syncytial virus

Respiratory syncytial virus is a common cause of lower respiratory tract disease and hospitalization in infants and young children [1]. Prematurity is one of the recognized risk

In the group of infants whose twin was not hospitalized in our center, a telephone survey was conducted as a follow-up to assess the condition of the second twin during this period (i.e., whether ill at home or hospitalized in a different center). The study was approved by the institutional ethics review board.

STATISTICAL ANALYSIS

Normally distributed data (e.g., age, birth weight) were expressed as mean \pm SD, non-normally distributed data as median and interquartile range, and categorical variables (e.g., gender) as percentages. For non-normally distributed data, comparisons were performed with the Mann-Whitney U test when appropriate, for normally distributed data with the independent-samples *t*-test, and for categorical data with the chi-square test. Univariate and multivariate logistic regression models were used to identify variables independently associated with the outcome variable, namely, the presence of severe RSV infection. Data were managed and analysed with the SPSS software package version 15.0 (SPSS, Chicago, IL, USA) and COMPARE 2.0 [7].

RESULTS

The study group included 875 infants with positive RSV antigen test, of whom 66 (7.6%) were of twin births. There were no triplets or higher order multiples during the study period. The characteristics of the two groups are shown in Table 1. Children in the twin group were younger than those in the singleton group (122.4 \pm 131.7 days compared to 204.5 \pm 278.8 days, $P = 0.014$) and had a lower mean gestational age (35.3 \pm 2.6 weeks vs. 38.6 \pm 2.5 weeks, $P < 0.001$). Infants in the twin group were more likely to have been born prematurely (< 37 gestational weeks) compared with the singleton group (65.6% vs. 13%, $P < 0.001$).

Table 2 shows the differences in the clinical course and treatment between twins and singletons with RSV-positive antigen test. The only significant difference between the groups was the presence of fever $\geq 38^\circ$ at admission ($P = 0.001$). While more infants in the twin group were admitted to the PICU and required mechanical ventilation, the differences between the groups did not reach statistical significance. Mean length of stay was also not significantly different between the two groups. On a multivariable logistic regression model, young age < 42 days (odds ratio 3.39, 95% confidence interval 1.46–7.9), early gestational age < 32 weeks (OR 10.58, 95%CI 3.25–34.54) and male gender (OR 1.97, 95%CI 1.05–3.69) were the only variables found as risk factors for PICU admission ($P < 0.001$, $P < 0.001$ and

PICU = pediatric intensive care unit
OR = odds ratio
CI = confidence interval

Table 1. Comparison of the demographic data between twins and singletons with RSV-positive antigen test

		Singletons (n=809)	Twins (n=66)	P value
Male/female (%)		57.2/42.8	53.1/46.9	0.453
Age group (days) Quintiles	< 42	18.9% (n=153)	25.8% (n=17)	0.02
	42–69	19.7% (n=159)	27.3% (n=18)	
	70–150	19.7% (n=159)	28.8% (n=19)	
	151–299	19.9% (n=161)	9.1% (n=6)	
	300–3720	21.9% (n=177)	9.1% (n=6)	
Gestational age (weeks)*	< 28	1.3% (n=10)	3% (n=2)	< 0.001
	29–32	1.8% (n=14)	6.1% (n=4)	
	33–37	17.1% (n=134)	77.3% (n=51)	
	> 38	79.8% (n=626)	13.6% (n=9)	
Chronic disease		9.4% (n=76)	4.5% (n=3)	0.3

*Data are missing for some infants

Table 2. Comparison of the clinical course and treatment between twins and singletons with RSV-positive antigen test

	Singletons (n=809)	Twins (n=66)	P value
Fever $\geq 38^\circ$ at admission	73.8% (n=597)	51.5% (n=34)	0.001
Oxygen treatment	72.1% (n=583)	72.7% (n=48)	1
Bronchodilator treatment	43.4% (n=351)	40.9% (n=27)	0.765
Steroid treatment	24.7% (n=200)	18.2% (n=12)	0.357
Antibiotic treatment	46.7% (n=378)	42.4% (n=28)	0.742
PICU admission	5.9% (n=48)	9.1% (n=6)	0.406
Mechanical ventilation	3.1% (n=25)	6.1% (n=4)	0.319
Mean length of stay (days)	5.76	5.38	0.69
Bacterial co-infection	2.6% (n=21)	4.5% (n=3)	0.556
Positive urine culture	2.0% (n=16)	4.5% (n=3)	0.325

$P = 0.03$, respectively). In contrast, the mere fact of a child being a twin was not found to be a significant risk factor on a multivariable logistic regression analysis.

A total of 53 pairs of twins were identified. In each pair, at least one of the twins was hospitalized at our center and was referred to as the “index case.” In 18 cases (34%) the other twin was also hospitalized during the same period with a clinical picture of bronchiolitis: 16 (30.2%) were hospitalized in our center (11 with a positive RSV antigen test, 5 with a negative RSV test) and 2 (3.8%) were hospitalized with a positive RSV antigen in other hospitals. Seventeen (32%) had respiratory complaints during the same time period but did not require hospitalization and 11 (20.8%) were symptom-free. One was still hospitalized in the neonatal intensive care unit due to prematurity during the same period and one died after birth. We had no information on 5 individuals (9%).

We compared the infants whose twin was also hospitalized during this period to infants whose twin was not hospital-

ized. The mean age of the index case twin was 66.7 days when both twins were hospitalized and 137.8 days when only one was hospitalized ($P < 0.001$). There was no significant difference between the two groups in other characteristics of the index case such as admission days, gestational week, birth weight and blood work parameters. On a multivariable model the age of the index case twin at admission was a significant risk factor ($P = 0.031$) for hospitalization of the second twin and each additional day lowered the risk of hospitalization for the second twin by 2%. Each additional week of gestation lowered the hospitalization risk for the second twin by 27%.

DISCUSSION

Our study demonstrated that during a 3 year period 7.6% of hospitalized infants with RSV infection were twins. The hospitalized infants in the twin group were younger than the singleton group ($P = 0.014$) and had a higher rate of prematurity ($P < 0.001$). In spite of these risk factors for severe RSV infection, and although a trend for more severe infection in the twin group was noted, this difference did not reach statistical significance; thus, according to our study multiple birth is not by itself associated with disease severity.

Simoes et al. [3] assessed the impact of multiple births on the severity of RSV infection in a retrospective cohort of preterm infants with bronchopulmonary dysplasia [3]. They found that the risk of developing RSV illness was significantly higher in multiple-birth infants than in singletons (53% vs. 24%, $P = 0.01$). They also found higher rates of RSV pneumonia among multiple-birth infants (24% vs. 6%, $P = 0.05$) [3]. However, their study focused on the subset of infants with chronic lung disease, and almost 20% of the infants in the multiple-birth group were triplets and not twins as in our study [3]. Resch and colleagues [4] retrospectively evaluated rates of rehospitalization due to respiratory illness in preterm infants of 29–36 weeks gestation without chronic lung disease. They found that multiple birth was associated with RSV-related hospitalization (55% vs. 15%, $P = 0.013$) [4]. However, it should be noted that these two studies included only a relatively small number of infants with RSV infection in the multiple birth group ($n=11-18$) [3,4]. In contrast, two larger prospective case-control studies, which were performed in Spain and evaluated risk factors linked to RSV infection requiring hospitalization in premature infants, found similar rates of infection in infants in the multiple-birth and in the control group [5,6].

Our study was not planned to answer the question of whether multiple birth is a risk factor for hospitalization due to RSV infection. Since we focused on infants who were already hospitalized, we tried to assess from a clinical point of view the risk for disease severity in this population and the risk for infection as well as hospitalization in the second

twin. Our data show that if one twin is hospitalized due to RSV infection, his/her twin has a 66% chance of acquiring a respiratory illness and a 34% chance of being hospitalized as well. The risk for hospitalization in the second twin was higher when the twin was younger in age. However, while more infants in the multiple-birth group were admitted to the PICU and required mechanical ventilation, the differences between the groups did not reach statistical significance. Although we studied 875 infants with RSV infection, a possible explanation for our findings may be the relatively small number of multiple-birth infants and the low rate of severe respiratory disease that required PICU admission.

The mechanism by which some infections, such as RSV illness, occur in multiple-birth infants and the severity of infections is determined partly by genetic factors as well as prenatal and postnatal environmental and sociodemographic factors [2]. Several studies recently hinted at the likely complexity and polygenic nature of genetic factors related to RSV severity. They identified certain specific chromosomal loci that possibly affect RSV disease severity, including those affecting the ability of the host to control early viral replication [8] and increased interleukin-13 production [9]. Previous studies also found higher risk of infection in twins from various bacterial as well as viral agents [10-14]. Twin gestation is a known risk factor for invasive Group B streptococcal infection, with a 25-fold greater likelihood of a twin of an affected infant acquiring infection compared to the same-age singleton population [11]. Moreover, susceptibility for various viral agents such as Epstein-Barr virus, enterovirus and human immunodeficiency virus were also found to be associated with both genetic and immunological factors that are concordant in twins [12-14].

Our study has several limitations. First, we used a retrospective design. Second, this study represents only the results of a single tertiary medical center, which accounted for the relatively small number of twins. A comprehensive prospective study collaborating local clinics, regional hospitals and medical centers will provide more details of RSV infection in the twin population. Third, we based our study on the ELISA test for RSV antigen. This test has some weaknesses: it is less sensitive than nucleic acid assays; it is also less sensitive than viral growth on tissue culture (alone or in combination with immunofluorescence) [15]. However, this method is still an acceptable alternative for the detection of the virus in respiratory secretions, and its reduced sensitivity (73%–93%) may have caused an underestimation of the total number of RSV infections but did not influence the proportion of infection in twins or the risk factors for its occurrence.

On the other hand, our study is the first to focus on the subgroup of hospitalized multiple-birth infants with RSV infection in order to identify risk factors for severe RSV infection. On

ELISA = enzyme-linked immunosorbent assay

a multivariable logistic regression model we found that age, low gestational age and male gender are risk factors for severe RSV disease. However, multiple-birth infants did not have an increased risk for developing severe RSV infection.

In summary, our retrospective investigation did not find a significantly higher rate of severe RSV infection in twin infants. However, we found high rates of hospitalization due to bronchiolitis or RSV infection in the second twin. These findings may help to identify those infants who are in a risk group for developing RSV infection that is severe enough to require close monitoring and hospitalization. Larger studies, perhaps on a national level, and more specific studies are needed to clarify the particular risk factors for RSV infection in multiple-birth infants.

Corresponding author:

M. Dotan

Dept. of Pediatrics A, Schneider Children's Medical Center of Israel, Petah Tikva 49202, Israel

Phone: (972-3) 925-3680

Fax: (972-3) 925-3958

email: miridotan@gmail.com, kontsy@gmail.com

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Capsule

Pathogenesis and transmission of avian influenza A (H7N9) virus in ferrets and mice

On 29 March 2013, the Chinese Center for Disease Control and Prevention confirmed the first reported case of human infection with an avian influenza A(H7N9) virus. The recent human infections with H7N9 virus, totaling over 130 cases with 39 fatalities to date, have been characterized by severe pulmonary disease and acute respiratory distress syndrome (ARDS). This is worrying because H7 viruses have typically been associated with ocular disease in humans, rather than severe respiratory disease. This recent outbreak underscores the need to better understand the pathogenesis and transmission of these viruses in mammals. Besler et al. assessed the ability of A/Anhui/1/2013 and A/Shanghai/1/2013 (H7N9) viruses, isolated from fatal human cases, to cause disease in mice and ferrets and to transmit to naive animals. Both H7N9 viruses replicated to higher titer

in human airway epithelial cells and in the respiratory tract of ferrets compared to a seasonal H3N2 virus. Moreover, the H7N9 viruses showed greater infectivity and lethality in mice compared to genetically related H7N9 and H9N2 viruses. The H7N9 viruses were readily transmitted to naive ferrets through direct contact but, unlike the seasonal H3N2 virus, did not transmit readily by respiratory droplets. The lack of efficient respiratory droplet transmission was corroborated by low receptor-binding specificity for human-like α 2,6-linked sialosides. These results indicate that H7N9 viruses have the capacity for efficient replication in mammals and human airway cells and highlight the need for continued public health surveillance of this emerging virus.

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Eitan Israeli

“The bitterest tears shed over graves are for words left unsaid and deeds left undone”

Harriet Beecher Stowe (1811-1896), American abolitionist and author. Her novel *Uncle Tom's Cabin* (1852) depicted the life of African Americans under slavery; it reached millions as a novel and play, and became influential in the USA and Britain. It energized anti-slavery forces in the American North, while provoking widespread anger in the South