Respiratory Syncytial Virus Infection: its Propensity for Bacterial Coinfection and Related Mortality in Elderly Adults

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Abstract

Respiratory syncytial virus (RSV) is an increasingly recognized cause of acute respiratory infection (ARI) in adults. We compared the crude in-hospital mortality of patients with RSV infection alone and with bacterial coinfection. Overall, 12,144 hospitalized patients with ARI were screened for RSV detection by PCR, between February 2014 and April 2019. In total, 701 (5.8%) had a positive RSV detection including 85 (12.1%) with bacterial coinfection. RSV-bacterial coinfection was associated with an increase in crude in-hospital mortality in patients over 65 years old (HR, 2.94 [95% CI, 1.30-6.60], P=.010). Optimized prevention and management strategies to reduce this burden are needed.

Keywords: Respiratory Syncytial Virus, Bacterial coinfection, Pneumonia, Mortality
Introduction

Lower respiratory infections are a major cause of mortality worldwide, with an estimated 2.38 million deaths in 2016 [1]. The four most common causative microorganisms are in order of frequency: *Streptococcus pneumoniae*, *Respiratory syncytial virus* (RSV), *Haemophilus influenza* type b, and Influenza viruses. Notably, this ranking is identical in children younger than 5 years and in elderly adults (>70 years). Regarding RSV, it is commonly responsible for bronchiolitis in children, whereas in adults it is associated with Influenza-like illness (ILI) or acute respiratory infections (ARI) including pneumonia. RSV represents an underestimated clinical burden in adults, being the causative agent of 10.6 to 13.5% of community pneumonia depending on whether patients are ambulatory or hospitalized [2,3]. Recent reports showed that in hospitalized patients with RSV-ARI, intensive care unit (ICU) admission was necessary in 15% to 16.8% of cases [4,5] and mortality ranged from 8% (in-hospital mortality) [5] to 11.9% at 60 days [6], being comparable to that associated with Influenza. In addition, recent reports showed that the hospital admission rate and in-hospital case-fatality ratio of RSV-ARI were higher for those aged ≥65 years than for those aged 50–64 years [7].

In adult hospitalized patients with RSV-ARI, bacterial-associated pneumonia was reported in from 42.3 to 79.7% of cases [5,6,8]. In children with bronchiolitis, positive blood culture or PCR on blood for the main respiratory bacterial pathogens were strongly associated to more severe RSV-infection [9]. In addition, the positive association between viruses and bacteria has been widely discussed in the literature, particularly for respiratory viruses and secondary bacterial pneumonia [10].

Herein we conducted a retrospective study over a five-year period in the four university hospitals of Marseille, France. We aimed to describe the epidemiology of bacterial coinfection in patients with RSV-ARI and compare crude in-hospital mortality of patients with RSV infection alone and with bacterial coinfection.
Material and methods

Study design

We conducted a retrospective historical cohort study of all adult patients hospitalized with RSV infection from 1st of February 2014 to 25th of April 2019 of the Assistance Publique – Hôpitaux de Marseille (AP-HM) in Marseille, France. Microbiological analysis was performed at the Institut Hospitalo-Universitaire (IHU) Méditerranée Infection laboratory which covers the four university hospitals of the institution.

Population

All patients aged 18 years or more hospitalized for ARI during the study period with an available result of RSV infection in absence of other respiratory virus infection during the same hospital stay were included. Hospitalization was defined as a length of stay ≥2 days. RSV requests with unavailable results and samples from outside the institution were excluded. Informed consent was not obtained from the patients as the study design was retrospective and did not modify standard of care.

Data collection

We screened all requests for RSV testing during the study period and used our local database to pair each RSV request with any bacteriological request made in our laboratory during the same patient’s hospital stay. The database included age, sex, date of admission, length of stay and in-hospital death for each patient. Microbiological characteristics included nature and date of collection of the samples, name and number of bacterial agents isolated. RSV infection was considered as nosocomial if the delay between admission and RSV-positive sample collection was equal or superior to 5 days (incubation period).

Virological and microbiological analysis

RSV positivity was defined either by a positive reverse transcriptase-polymerase chain reaction (RT-PCR) specific for RSV with a cycle threshold (Ct) value ≤35 cycles or by a positive immunochromatographic test specific for RSV antigen on any of the following samples: pharyngeal
swab, nasal swab, nasopharyngeal aspirate, sputum or bronchoalveolar lavage (BAL). Syndromic and point-of-care culture and culture-independent testing including viral and bacterial detection were requested by clinicians for patients presenting ARI. Bacterial coinfection was defined as the culture of a known respiratory pathogen from blood and/or a respiratory sample (sputum, bronchial aspiration or bronchoalveolar lavage), or a positive culture-independent test including *Streptococcus pneumoniae* or *Legionella pneumophila* antigen detection in urine, or *Mycoplasma pneumoniae* or *Bordetella pertussis* PCR, less than two days before or fourteen days after the RSV-positive test. In patients with ARI, a syndromic point-of-care testing was systematically performed. Patients with a blood culture or a respiratory sample positive for coagulase-negative *Staphylococcus* (or other bacteria without known pulmonary pathogenicity) were not considered as coinfected.

**Statistical analysis**

Two-group comparisons were performed with Student test (or Mann-Whitney test when appropriate) for quantitative variables and with Fisher’s exact test (or Chi-squared test when appropriate), for qualitative variables. Survival analyses were performed using Kaplan-Meier curve with log-rank test and Cox model regression (multivariate analysis). A significance threshold of 0.05 was adopted for all statistical analyses. Statistical analysis was performed with PASW Statistics version 17.0.
Results

Population

During the study period, we recorded 50,921 requests for RSV testing. 2,080 requests (4.1%) had unavailable RSV results and were excluded. A total of 10,155 requests (19.9%) were made during ambulatory stay in hospital and were excluded. The remaining 38,686 requests represented 26,811 in-hospital patients and among them 12,144 (45.2%) were included (See Supplementary Figure 1). We excluded patients positive for influenza A or B viruses, rhinovirus, adenovirus or metapneumovirus during the same hospital stay. In total, 11,443 (94.3%) patients were RSV-negative and 701 (5.7%) were RSV-positive; 291 (45.1%) were male. Mean age of RSV-positive patients was 69.9±19.3 years (range, 60-85), and 483 patients (68.9%) were older than 65 years. Global in-hospital mortality rate was 1.4% during the studied period. The mean age of deceased patients was 80.6±8.8 years (range, 73.5-87.5). A bacterial coinfection was found in 85 (12.1%) patients with RSV-ARI. Age, sex, length of hospital stay and rates of RSV nosocomial acquisition did not differ according to the presence of bacterial coinfection. The crude in-hospital mortality was significantly higher in patients with RSV and bacterial coinfection when compared to RSV-ARI alone (P=.01) (Table 1).

Microbiological analysis

Among the 95 bacteria associated with RSV-ARI, 64 (67.4%) were identified by culture (blood culture: 22 (34.3%); respiratory samples: 42 (65.7%)), 26 (27.4%) by culture-independent methods and 5 (5.2%) by both techniques.

Streptococcus pneumoniae was the most frequently identified bacteria (23.5%), followed by Mycoplasma pneumoniae (14.1%), Pseudomonas aeruginosa (11.8%), and Staphylococcus aureus (10.6%). The bacteria isolated in at least three patients are shown in Supplementary Figure 2.
Mortality and bacterial coinfection in adult patients with RSV infection

The majority of deaths (97.5%) occurred in patients over 65 years old (see Supplementary Figure 3). We therefore performed a survival analysis in this specific age class. In patients ≥65 years, survival univariate analysis showed that RSV-bacterial coinfection was associated with higher mortality rate (18.8% vs. 7.0%; HR 2.7 [1.2-6.1]; log-rank test, \( P = .010 \)). This finding remained unchanged after adjustment for age and length of stay in multivariate analyses (Cox model regression, HR, 2.94 [95% CI, 1.30-6.60], \( P = .010 \)) (Figure 1).

Discussion

In this retrospective study over a five-year period, we found that 12.1% of hospitalized patients with RSV-ARI had bacterial coinfection. This is lower than the rate reported by Jung et al. (19.7%) [11], but higher than that reported by Jeannoël et al. (9.3%) [12]. The latter study analyzed exclusively RSV-positive pneumonia whereas we analyzed all RSV-positive ARI. This may have contributed to differences in the frequency of associated bacteria across studies as they reported in descending order *Streptococcus pneumonia*, *Haemophilus influenza*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Moraxella catarrhalis*. These ecological differences may also be partly explained by the used microbiological tests. Indeed, the systematic testing in our syndromic point-of-care testing included PCR specific for *M. pneumoniae* in patients with ARI.

Although the bacterial coinfection burden and outcome have been widely described in patients with viral respiratory infections [13], few studies analyzed it in patients with RSV-ARI. Recently, Jeannoël et al. observed a more severe outcome for RSV plus bacteria-associated pneumonia compared to only RSV-associated pneumonia. In particular, the length of hospital stay was significantly longer and ICU hospitalization, more frequent. Although difference in hospitalization characteristics was not observed in the present study, we found that bacterial coinfection was associated with higher mortality rate in patients over 65 years old with RSV-ARI. This finding remained unchanged after adjustment for age and length of stay in multivariate analysis. As RSV-ARI...
and bacterial coinfection may result in greater morbidity and mortality among older hospitalized adults, vaccination and early antibiotic treatment strategies need to be further evaluated and implemented in this population [3,14]. Moreover, clinicians should perform exhaustive and systematic detection of RSV and bacterial coinfections in hospitalized adult patients with ARI, particularly in elderly subjects.

Our findings suffer from some limitations. First, the retrospective design exposes to biases. Second, lack of clinical data prevents accurate diagnosis of ARI subtype, and analysis of factors such as illness severity, comorbidities, and timing of appropriate antibiotic administration. Third, outcome was only available during in-hospitality stay. Finally, the causal role of RSV infections in the clinical presentation and outcome would be better assessed in a case-control study. Nonetheless, asymptomatic RSV infection is uncommon in elderly and a recent meta-analysis suggests that there is strong evidence for the association between RSV and ARI [15]. Notably, among individuals with RSV-ARI, a causative role for ARI was attributable to RSV in about 88% of cases.
Figure Legends

Figure 1. Effect of bacterial co-infection on crude in-hospital mortality in RSV-positive hospitalized patients aged over 65 years old.

Green line: RSV infection alone; Blue line: RSV infection with bacterial coinfection. Kaplan-Meier curve, log-rank test, \( P = .010 \).

RSV = *Respiratory Syncytial Virus*
Compliance with ethical standards

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Conflicts of interest: All authors report no conflicts of interest relevant to this article.

Patient Consent Statement: This study does not include factors necessitating patient consent. The design of the work has been approved by our local ethical committee.
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Table 1. Comparison of epidemiological and clinical features among RSV-infected patients in absence or presence of bacterial co-infection

Sd=Standard Deviation; RSV=Respiratory Syncytial Virus

|                              | RSV alone (N = 616) | Bacterial co-infection (N = 85) | P     |
|------------------------------|---------------------|---------------------------------|-------|
| Age (mean ± sd)              | 70.4 ± 19.4         | 66.6 ± 18.6                     | 0.083 |
| Women (n (%))                | 367 (59.6)          | 43 (50.6)                       | 0.098 |
| Length of stay (days, mean ± sd) | 9.2 ± 7.1        | 9.6 ± 6.2                       | 0.58  |
| Hospital-acquired RSV (n (%)) | 101 (16.4)         | 15 (17.6)                       | 0.75  |
| In-hospital mortality (n (%)) | 30 (4.9)            | 11 (12.9)                       | 0.01  |
Figure 1

Cumulative survival

Time from inclusion (days)

COinfected RSV | 45 | 43 | 40 | 38 | 37 | 37 | 37
Non-COinfected RSV | 358 | 349 | 341 | 338 | 337 | 336 | 334