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Mortality Rates of Human Metapneumovirus and Respiratory Syncytial Virus Lower Respiratory Tract Infections in Hematopoietic Cell Transplantation Recipients

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A B S T R A C T

Human metapneumovirus (HMPV), a common respiratory virus, can cause severe disease in pre- and post-hematopoietic cell transplantation (HCT) recipients. We conducted a retrospective cohort analysis in HCT patients with HMPV (n = 23) or respiratory syncytial virus (n = 23) detected in bronchoalveolar lavage samples by reverse transcription PCR between 2006 and 2011 to determine disease characteristics and factors associated with outcome. Mortality rates at 100 days were 43% for both HMPV and respiratory syncytial virus lower respiratory tract disease. Steroid therapy, oxygen requirement >2 L or mechanical ventilation, and bone marrow as cell source were significant risk factors for overall and virus-related mortality in multivariable models, whereas the virus type was not. The presence of centrilobular/nodular radiographic infiltrates was a possible protective factor for mechanical ventilation. Thus, HMPV lower respiratory tract disease is associated with high mortality in HCT recipients. Earlier detection in combination with new antiviral therapy is needed to reduce mortality among HCT recipients.

INTRODUCTION

Human metapneumovirus (HMPV) is a paramyxovirus closely related to respiratory syncytial virus (RSV). HMPV occurs with a seasonal pattern in the general population every winter and spring. Both HMPV and RSV cause similar symptoms in immunocompetent children and adults and are impossible to distinguish on a clinical basis [1]. HMPV infections can cause severe and even fatal disease in immunocompromised patients [2,3], with crude reported mortality rates from HMPV pneumonia ranging from 10% to 80% in different small cohort studies of cancer and/or hematopoietic cell transplantation (HCT) patients [3-6]. To date, however, no study has directly compared lower respiratory tract disease (LRTD) associated with HMPV with that of RSV in immunocompromised patients. Although the treatment and outcome of RSV LRTD in HCT has been relatively well studied and standardized [7,8], few data are available on the impact of HMPV LRTD during the time surrounding HCT. The purpose of this study was to characterize the clinical and radiographic presentation, viral load, and factors associated with outcome of HMPV pneumonia in HCT candidates and recipients and compare results with RSV pneumonia.

METHODS

Patients and Samples

We retrospectively reviewed medical charts of all pre- and post-HCT recipients with HMPV or RSV RNA detected in bronchoalveolar lavage (BAL) samples by real-time reverse transcription (RT)-PCR. Molecular analyses on BAL were done clinically in real time starting in January 2006 for HMPV and March 2007 for RSV. This study included patients undergoing BAL with HMPV or RSV detected through February 1, 2011. The RSV cases are a subset of a series reported earlier [9]. Sera and nasal-wash specimens that had been collected 11 days before to 11 days after each BAL were retrospectively identified and, if not previously tested for these viruses, were evaluated for the presence of HMPV and RSV by RT-PCR.

Research was approved by the Institutional Review Board at Fred Hutchinson Cancer Research Center. Informed consent was signed by study participants.

Laboratory Method

In addition to RSV and HMPV RT-PCR, respiratory viral diagnosis for multiple respiratory viruses was performed on BAL specimens from pre- and post-HCT recipients according to institutional protocol. Direct fluorescent antibody (DFA) testing for influenza A and B, parainfluenza 1-3, adenovirus, and RSV as well as RSV shell vial cultures were performed on BAL samples between 2006 and 2011 as described in a previously published protocol [10]. HMPV DFA testing was performed on BAL specimens from February 2008 through 2011. BAL samples were also assessed for a broad range of bacterial, fungal, and viral pathogens using standard culture and staining methods as well as the aspergillus galactomannan assay [11,12].

RT-PCR was performed on BAL, nasal-wash, and sera specimens according to a previously published protocol [13]. Briefly, total nucleic acids were obtained from 200 μL of each BAL or nasal-wash sample by adding 400 μL lysis buffer. After incubation for 10 minutes at 60°C, 600 μL of isopropanol was added and the samples were centrifuged at 13,000 × *g* for 15 minutes. The pellets were washed with 1 mL 70% ethanol and suspended in 200 μL RNAse free water. One-step RT-PCR reaction mixtures (TaqMan One-Step RT-PCR Master Mix, Applied Biosystems, Foster City, CA) were prepared using primers and probes targeting HMPV A/B and an internal control or RSV A/B and an internal control as previously published [13]. The reactions were performed and analyzed in a 7000 Sequence Detection System (Applied Biosystems) under the following conditions: 30 minutes at 48°C and 10 minutes at 95°C, followed by 40 cycles of 15 seconds at 95°C and 1 minute at 60°C. Sera extractions were performed using the QIAamp RNA Mini Kit (Valencia, CA) as previously reported and provided a sensitivity of 200 copies/mL for a cut-off of 10 copies per reaction [14]. Cycle threshold values were converted into viral loads (copies/mL) of each virus using stored standard curves made of RNA transcripts.

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Statistical Methods

Statistical comparisons were performed using chi-square test or Fisher's exact test for categorical variables (as appropriate), and Wilcoxon rank sum test was used for continuous variables. The probability of survival was estimated by the Kaplan-Meier method. Univariate and multivariate logistic regression models were used to estimate odds ratios (ORs) and 95% confidence intervals (CIs) for hypoxemia. Univariate and multivariate Cox regression models were used to evaluate hazard ratios (HRs) and 95% CIs for death by day 100 and virus-related death at day 100. Hypoxemia and mechanical ventilation occurring after diagnosis were analyzed as time-dependent variables. All reported *P* values were 2-sided and considered significant if $P < .05$. Analyses were performed using SAS 9.2 (SAS Institute, Cary, NC).

RESULTS

Patients

Between January 2006 and February 2011, 23 severely immunocompromised pre- or post-HCT patients had HMPV detected by RT-PCR from BAL specimens. Between March 2007 and February 2011, 23 separate patients were found to have RSV LRTD. Both groups had similar demographic characteristics and levels of immunosuppression (Table 1). All patients infected with RSV (with the exception of one who declined therapy) were treated with aerosolized ribavirin and palivizumab according to our institutional protocol; treatment for HMPV was nonstandardized and differed by care provided, because no standardized institutional treatment guidelines were available. Among the 23 patients with HMPV LRTD, four received ribavirin alone, five received intravenous immunoglobulin alone, and six received both. Time to treatment after first positive respiratory sample (including nasopharyngeal secretions or BAL) was a median of 5 days (range, 1 to 46 days) for HMPV and 1 day (range, 0 to 4 days) for RSV (Wilcoxon rank; $P = .0005$), and time to treatment after first positive BAL had a median of 2 days (range, −13 to 7 days) for HMPV and 0 days (range, −15 to 2 days) for RSV (Wilcoxon rank; $P = .009$).

HMPV and RSV were generally detected between December and June. One RSV cluster occurred during the study period, December 2007, in which BAL samples from seven

Table 1
Demographic and Clinical Variables of Patients with Positive RT-PCR for HMPV or RSV from BAL Samples

Demographics and Clinical Variables	RSV (n = 23)	HMPV (n = 23)	*P*
Sex, male	16 (70)	15 (65)	.75
Age, yr, median (IQR)	58 (46-67)	50 (32-63)	.095
HSCT (type)			
Autologous	4 (17)	3 (13)	.84
Allogeneic	15 (66)	14 (61)	
Pre-HSCT	4 (17)	6 (26)	
Cell source			
Bone marrow	3 (13)	6 (26)	.43
Peripheral blood stem cell	18 (78)	13 (57)	
Cord blood	2 (9)	3 (13)	
No transplantation	0	1 (4)	
Total body irradiation*			
12 Gy	3 (16)	2 (12)	.58
2 Gy	9 (47)	11 (65)	
None	7 (37)	4 (24)	
GVHD†	13 (81)	11 (79)	1
Time after HCT,* days, median (IQR)	106 (17-271)	80 (20-361)	.85
Time after HCT >100 days*	10 (53)	8 (47)	.74
Lymphocyte count under 300 cells/μL at time of BAL	9 (39)	11 (48)	.55
CMV reactivation within 1 mo before BAL*	7 (37)	6 (35)	.92
Steroids within 2 wks before BAL	13 (57)	9 (39)	.24
Copathogen‡	8 (35)§	13 (57)‖	.18
Oxygen at diagnosis (>2 L + ventilation)	11 (48)	8 (35)	.23
Radiologic variables¶			
Centrilobular/nodular	13 (59%)	8 (36%)	.13
Ground glass	15 (68%)	13 (59%)	.53
Tree-in-bud	4 (18%)	4 (18%)	1
Alveolar	14 (64%)	15 (68%)	.75
Treatment variables			
Treatment with ribavirin only	0 (0)	4 (17)	NA
Treatment with ribavirin and IVIG/palivizumab	22 (96)	6 (26)	NA
Treatment with IVIG only	0 (0)	5 (22)	NA
Time to start of treatment from first positive sample, days, median (IQR)	1 (1-1)	5 (2-7)	.0005
Time to start of treatment from first positive BAL, days, median (IQR)	0 (−1-1)	2 (1-5)	.009
Outcomes			
Hypoxemia	16 (70%)	15 (65%)	.75
Mechanical ventilation	10 (43%)	7 (30%)	.36
Death at 100 days	10 (43%)	10 (43%)	1
Death related to RSV or HMPV infection	8 (35%)	9 (39%)	.76

IQR indicates interquartile ratio; CMV, cytomegalovirus; GVHD, graft-versus-host disease; IVIG, intravenous immunoglobulin.
Values are total number of incidences with percents in parentheses, unless otherwise noted.

* HCT recipients only.

† Allo HCT recipients only.

‡ Some patients had more than one pathogen identified in the BAL or in the blood at presentation. When another respiratory virus was identified, RSV or HMPV was always the predominant virus based on viral loads. Fungal infections were determined by blood or BAL galactomannan, microbiology, or pathology and most were known and treated at the time of respiratory virus diagnosis.

§ Fungal (3), *S. aureus, enterococcus* (3), *E. coli, Klebsiella* (2), rhinovirus (2), *S. viridians* sepsis, *Klebsiella* sepsis.

‖ *Pneumocystis jiroveci*, fungal (9), *Haemophilus, S. pneumonia, Stenotrophomonas,* enterococcus, parainfluenza, influenza A, rhinovirus (3), coronavirus (3), CMV in BAL (2), herpes simplex virus in BAL, enterococcus sepsis, staphylococcus sepsis, gram-negative rod sepsis, vancomycin-resistant enterococcal sepsis.

¶ Only 22 of 23 patients with HMPV LRTD and 22 of 23 patients with RSV LRTD had axial tomography performed.