Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.
Viral Respiratory Tract Infections in Allogeneic Hematopoietic Stem Cell Transplantation Recipients in the Era of Molecular Testing

Starling A. Sim 1,2, Vivian K.Y. Leung 1,3, David Ritchie 2,4, Monica A. Slavin 2,3,5,6, Sheena G. Sullivan 1,7, Benjamin W. Teh 3,5,6,*

1 World Health Organization Collaborating Centre for Reference and Research on Influenza, Doherty Institute for Infection and Immunity, Melbourne, Victoria, Australia
2 Department of Medicine, University of Melbourne, Melbourne, Victoria, Australia
3 Department of Infectious Diseases, Peter MacCallum Cancer Centre, Melbourne, Victoria, Australia
4 Department of Haematology, Royal Melbourne Hospital, Melbourne, Victoria, Australia
5 National Centre for Infections in Cancer, Melbourne, Victoria, Australia
6 Sir Peter MacCallum Department of Oncology, University of Melbourne, Melbourne, Victoria, Australia
7 School of Population and Global Health, University of Melbourne, Melbourne, Victoria, Australia

Article history:
Received 5 December 2017
Accepted 5 March 2018

Key Words:
Respiratory virus
Allogeneic
Risk factors
Outcomes

ABSTRACT
Viral respiratory tract infection (vRTI) is a significant cause of morbidity and mortality in patients undergoing allogeneic hematopoietic stem cell transplantation (allo-HSCT). This study aimed to assess the epidemiologic characteristics, risk factors, and outcomes of vRTI occurring in the period from conditioning to 100 days after allo-HSCT in the era of molecular testing. This study was a retrospective record review of patients who underwent allo-HSCT at Royal Melbourne Hospital between January 2010 and December 2015. Symptomatic patients were tested using respiratory multiplex polymerase chain reaction (PCR). Logistic regression and Kaplan-Meier analysis were used to identify risk factors for vRTI and the risk of death or intensive care unit (ICU) admission, respectively. A total of 382 patients were reviewed, and 65 episodes of vRTI were identified in 56 patients (14.7%). Rhinovirus accounted for the majority of infections (69.2%). The majority of episodes presented initially with upper respiratory tract infection (58.5%), with 28.9% of them progressing to lower respiratory tract infection. Eleven episodes (16.9%) were associated with ICU admission. There were no deaths directly due to vRTI. Previous autologous HSCT was associated with an increased risk of vRTI (odds ratio, 2.1; 95% confidence interval, 1.0 to 4.1). The risks of death (P = .47) or ICU admission (P = .65) were not significantly different by vRTI status. vRTI is common in the first 100 days after allo-HSCT and is associated with ICU admission. © 2018 American Society for Blood and Marrow Transplantation.

INTRODUCTION
Background
Respiratory viruses (RVs), including influenza virus, parainfluenza virus, respiratory syncytial virus (RSV), rhinovirus, coronavirus, adenovirus, and human metapneumovirus (hMPV), are common causes of viral respiratory tract infection (vRTI) [1-3]. Previous studies have shown that RVs cause significant morbidity and mortality in patients undergoing hematopoietic stem cell transplantation (HSCT), in particular allogeneic HSCT (allo-HSCT) [1-8].

The diagnosis of RVs, especially in older studies, has been dependent mainly on nonmolecular diagnostic methods, including direct antigen detection or cell culture, which are limited by poor sensitivity [9]. Molecular testing using polymerase chain reaction (PCR) allows rapid detection of RVs with high sensitivity and specificity. Moreover, multiplex assays enable the detection of multiple RVs in a single test, and these are now the current standard of care in many clinical settings. The purpose of this study was to assess the epidemiologic characteristics, risk factors, and outcomes of vRTI in allo-HSCT recipients in the era of molecular testing.

METHODS
Study Population
This study was a retrospective record review of adult patients admitted to the Bone Marrow Transplantation (BMT) unit for allo-HSCT at the Royal...
Melbourne Hospital (RMH) between January 2010 and December 2015. Only the first allo-HSCT was considered in patients who underwent multiple transplantations during the study period.

Patient records for the first 100 days following allo-HSCT were reviewed. Demographic and clinical data were collected from hospital clinical records using a case report form and included age, sex, underlying disease, previous therapy, stem cell source, conditioning therapy, graft-versus-host disease (GVHD), and outcomes (ie intensive care unit [ICU] admission and death). Baseline measurements of lymphocytes and neutrophil levels and cytomegalovirus (CMV) seropositivity were collected. Data on respiratory viral PCR testing and results from symptomatic patients were extracted from the RMH pathology database.

For patients with RV infection, the number of vRTIs, type of RVs, clinical presentation, antiviral therapy, and outcomes (ie, ICU admission, death, use of mechanical ventilation, and progression to lower respiratory tract infection [LRTI]) were also obtained during the 100-day period. During the review period, treatment of RV with i.v. ribavirin was recommended in the setting of radiologic changes in patients within 100 days of allo-HSCT.

**Microbiological Methods**

All specimens, including nasal and throat swabs, nasopharyngeal aspirates, and sputum or bronchoalveolar lavage specimens, were examined by real-time PCR at the Melbourne Health Shared Pathology Service according to previously published protocols [10,11]. RVs included in the multiplex PCR panel were RSV, influenza type A and B, and rhinovirus. The testing of copathogens was not part of routine assessment and was performed at the discretion of treating physician. These tests included conventional culture for bacteria and fungi; PCR for Legionella, Pneumocystis jirovecii, and Aspergillus [12]; viral PCR for herpes simplex virus (HSV)-1, HSV-2, CMV, varicella zoster virus, human herpesvirus (HHV)-6, HHV-8, HPV, and adenovirus; and bacterial PCR for Mycoplasma pneumoniae, Chlamydia phila pneumoniae, and Chlamydia psittaci.

**Definitions**

Baseline measurements of lymphocytes and neutrophils were defined as measurements obtained on the day of admission ± 2 days. Lymphopenia was defined as an absolute lymphocyte count below 1.0 x 10^9/L, and neutropenia was defined as an absolute neutrophil count below 2.0 x 10^9/L. Coughing on admission was defined in accordance with Center for Additional Blood and Marrow Transplant Research guidelines [13]. Upper respiratory tract infection (URTI) was defined as an RV detected in an upper respiratory tract fluid specimen together with symptoms and/or signs, with the exclusion of other possible causes. LRTI was defined as detection of RV in respiratory secretions, preferentially in samples obtained from sites of involvement together with pathologic suprathymic production, hypoxia, or pulmonary infiltrates [14]. Progression to LRTI was defined as the onset of LRTI in patients with a previous URTI. An episode of infection was defined as evidence of RV, URTI, or LRTI detected during the conditioning period and for up to 100 days following allo-HSCT. An episode of infection was defined as evidence of RV, URTI, or LRTI detected during the review period. An episode of LRTI with rhinovirus infection (n = 160; 41.9%) was defined as the onset of LRTI in patients with a previous URTI. An episode of LRTI was defined as death due to any cause within 100 days of vRTI diagnosis or allo-HSCT. Death attributable to vRTI was defined as death resulting from respiratory failure with other causes excluded.

**Statistical Methods**

The risk of vRTI was calculated as the number of patients testing positive for any RV among all allo-HSCT recipients. Patients’ baseline characteristics were compared by vRTI status using the chi-square test or Fisher exact test for categorical variables and the Student t test or Wilcoxon rank-sum test for continuous variables.

Only the first episode of vRTI was considered for risk factor analysis. Univariable and multivariable analyses of risk factors for vRTI were performed using logistic regression models. Variables with a P value < .1 were included in the multivariable testing, along with variables previously identified as important risk factors for vRTI, including lymphopenia, GVHD, and donor relation [16,17].

Patients with vRTI had their records reviewed for 100 days following infection. In patients without vRTI, the review was censored to the date of death/ICU admission or 100 days following transplantation, whichever was sooner. Survival (to death or ICU admission) within 100 days following transplantation was assessed using Kaplan-Meier plots, stratified by vRTI status. The log-rank test was used to assess differences in survival among groups. All statistical analyses were performed using Stata-13 (3.3.1), with statistical significance defined at α = .05.

**RESULTS**

**Patient Characteristics**

A total of 386 patients were identified during the review period (January 2010 to December 2015). Medical records could not be obtained for 4 patients, including 1 patient with vRTI (Figure 1). The 382 patients reviewed included slightly more males (n = 222; 58.1%) than females (n = 160; 41.9%), and the median patient age was 47 years (interquartile range [IQR], 36 to 56 years). The median age of patients with vRTI was significantly lower than patients without (P = .03). Other patient demographic data and characteristics were not significantly different by vRTI status (Table 1).

**Prevalence of vRTI**

Out of 382 patients, 222 were tested for vRTI (58.1%) within 100 days following allo-HSCT and 65 episodes of vRTI were identified in 56 patients (14.7%). Eight patients had multiple episodes, 7 with 2 episodes and 1 with 3 episodes. The median time to onset of the first infection from the date of allo-HSCT was 41 days (IQR, 9.8 to 64.0 days). Among the 56 patients identified with vRTI, their first episode was identified during the first 30 days following transplantation in 23 (41.1%). The clinical characteristics of vRTI episodes are summarized in Table 2.

Rhinovirus accounted for the majority of infection episodes (45 of 65; 69.2%), followed by RSV (11 of 65; 16.9%), and there was 1 episode of coinfection with these 2 pathogens. Most of the cases initially presented with URTI (38 of 65; 58.5%), and this was most pronounced for RSV (8 of 11; 72.7%). Progression to LRTI was observed in 28.9% (11 of 38) of all patients with an initial presentation of URTI and was most common for rhinovirus (8 of 25; 32.0%).

The most common symptom observed was cough (n = 51; 78.5%), followed by fever (26; 40.0%), corony (23; 35.4%), and sputum production (19; 29.2%). The rates of cough (100% versus 68.4%), sputum production (76.0% versus 0%), and fever (60.0% versus 29.0%) were significantly higher in the patients with LRTI compared with those with URTI (all P < .05). Antiviral therapy was administered in 14 cases (21.5%). Among the 11 patients with RSV, 3 (27.3%; 1 URTI and 2 LRTI) received ribavirin, and 1 patient with LRTI received either placebo or gs-5806 as part of a clinical trial. All 9 patients who had either influenza A or B received oseltamivir, and 1 patient with an LRTI with rhinovirus infection (n = 1/45; 2.2%) received i.v. immunoglobulin. Antiviral therapy was initiated within a median of 2.0 days (IQR, 1.0 to 2.8 days) after the detection of RV. The median duration of antiviral therapy was 5.5 days (IQR, 4.0 to 8.8 days).

Sixteen copathogens were identified from 10 vRTI episodes (15.4%). One episode had 4 copathogens (3 bacterial and 1 fungal), and 3 episodes had 2 copathogens (2 episodes with 2 fungal pathogens and 1 episode with a bacterial and a fungal pathogen). Bacterial copathogens were found in 8 out of 10 episodes (80.0%) and accounted for one-half the copathogens detected (8 of 16; 50.0%). These included Staphylococcus spp. (n = 3), Enterococcus faecium (n = 2), Haemophilus influenzae (n = 1), Chlamydia pneumoniae (n = 1), and Pseudomonas aeruginosa (n = 1). Of note, fungal and nonrespiratory virus copathogens were only found concomitantly with rhinovirus infection. Fungal copathogens identified include Aspergillus (n = 2), Candida albicans (n = 1), Cladosporium sp.
(n = 1), Saccharomyces cervisiae (n = 1), and P. jirovecii (n = 1). No episodes met the European Organization for Research and Treatment of Cancer/Mycoses Study Group criteria for invasive fungal disease. The nonrespiratory viruses isolated were CMV and HHV-6.

Risk Factors for vRTI

In univariate and multivariable analyses, only previous autologous HSCT was associated with increased risk of vRTI (odds ratio, 2.1; 95% confidence interval, 1.0 to 4.1). Other variables, including age, sex, underlying disease, CMV seropositivity, donor relation, stem cell source, conditioning regimen and intensity, acute GVHD, lymphopenia, and neutropenia were not statistically significant (Table 3). In addition, no variables were significantly associated with the risk for vRTI for fixed periods following HSCT (<30 days, 30 to 60 days, or 61 to 100 days following transplantation).

Outcomes of vRTI

There were a total of 65 episodes of vRTI in our study cohort. During the study period, 11 episodes (16.9%) were associated with ICU admission within 100 days of vRTI, with 9 episodes (13.8%) necessitating the use of mechanical ventilation. Of the 11 episodes, 5 episodes of vRTI (2 rhinovirus, 2 RSV, and 1 influenza A) had LRTI as the initial presentation and respiratory symptoms that precipitated the ICU admission. Patients were admitted to ICU within a median of 6.0 days (IQR, 0 to 15.5 days) after vRTI and for a median duration of 9 days (IQR, 5.5 to 12.5 days). The risk of ICU admission within 100 days following transplantation between patients with vRTI (9 of 56; 16.0%) and those without vRTI (44 of 326; 13.5%) was not significantly different (P = .65) (Figure 2).

Five patients died within 100 days of vRTI, all of whom had rhinovirus infection and prior ICU admission. Three patients were admitted to the ICU 1 day before the detection of rhinovirus, whereas the other 2 patients had ICU admission on day 22 and day 38 following the detection of rhinovirus. Of the 5 patients who died, 4 (80.0%) had LRTI as the initial site of infection and 3 (60.0%) had copathogens detected. However, none of these deaths was directly attributable to vRTI. Causes of death included multiorgan failure (n = 2), pneumonitis (n = 1), acute pulmonary edema (n = 1), and aspiration pneumonia, GVHD, and fluid overload (n = 1). The risk of mortality within 100 days following transplantation between patients with vRTI (3 of 56; 5.4%) and those without vRTI (27 of 326; 8.3%) was not significantly different (P = .47) (Figure 2). All vRTI patients who died within 100 days following transplantation developed vRTI during the first 30 days following transplantation.

DISCUSSION

In this study, vRTI was prevalent among allo-HSCT recipients during the first 100 days following transplantation. The frequency of vRTI among allo-HSCT recipients was 14.6%, which is consistent with the rates of 3.5% to 29% reported elsewhere [2,3,6,7]. However, differences between our study and previous studies include a shorter duration of review (100 days), inclusion of symptomatic patients only, and the range of pathogens tested [2,3,6,7,17].
Rhinovirus accounted for the majority of infections, as has been reported previously, albeit among symptomatic and asymptomatic patients [2,17]. Copathogens were identified in 15% of the episodes and were most common in patients with rhinovirus infection. Ison et al. [18] hypothesized that rhinovirus might predispose patients to additional infection. Respiratory colonization with potentially pathogenic bacteria also may increase the risk of subsequent vRTI [19]. However, we note that in both that study and our present study, the number of patients was small, and further investigation of a potential link is warranted.

In our cohort, patients who had undergone previous auto-HSCT were at increased risk of developing vRTI independent of GVHD, donor relation, and lymphopenia. Auto-HSCT has been identified as an important risk factor for progression to LRTI [8]. Previous auto-HSCT may reflect advanced disease status and thus the susceptibility of patients to vRTI. In addition, cumulative immune suppression from multiple lines of previous therapy may increase the risk of vRTI, as has been seen in patients with multiple myeloma [15]. Delays in immune recovery following auto-HSCT also could explain the increased risk of vRTI in this patient population [20,21].

A key finding of the present study was the low number of deaths in patients with vRTI, with no mortality directly attributable to vRTI. This finding was consistent with rates reported in previous studies (0% to 4.7%) [3,8,22,23]. We were unable to directly assess factors contributing to reduced mortality, but postulate that it may be attributable to high standards of supportive care. In addition, antiviral therapy was initiated in some RSV cases and in all episodes of influenza type A and B infection early in the course of infection. Among the patients with vRTI, all deaths occurred in patients who acquired the infection within 30 days of transplantation. This high risk and poor prognosis subgroup supports the need for strict infection prevention measures for staff and patients in the early days after transplantation [17,23-27], along with careful assessment of patients presenting for allo-HSCT with respiratory symptoms.

Our study demonstrates that vRTI is an important cause of morbidity in allo-HSCT recipients. Up to 17% of vRTI episodes necessitated ICU admission and 14% required the use of mechanical ventilation, percentages consistent with previous reports [24-26]. Overall, close to 40% of patients had LRTI on initial presentation. Progression to LRTI occurred in 31% of infections, most commonly involving rhinovirus. Progression to LRTI was also observed in 25% of RSV URTI episodes, a higher rate than reported previously [28]. Both the innate and adaptive arms of the immune system, particularly neutrophils and cytotoxic T cells, play integral roles in the defense against vRTI [19]. Th1-dominant responses,

---

Table 1: Characteristics of Allo-HSCT Recipients with and without vRTI

| Variable                        | vRTI   | Non-vRTI | P Value |
|---------------------------------|--------|----------|---------|
| Number of patients (%)          | 56 (14.7) | 326 (85.3) |         |
| Age, yr, median (range)         | 41.9 (18.7-61.5) | 47.5 (17.0-69.6) | .03*    |
| Sex, n (%)                      | 19 (33.9) | 141 (43.3) | .25     |
| Female                          | 37 (66.1) | 185 (56.7) |         |
| Male                            |        |          |         |
| Underlying disease, n (%)       |        |          |         |
| Malignant                       | 55 (98.2) | 312 (95.7) | .71†    |
| AML                             | 7 (12.5) | 40 (12.3) |         |
| ALL                             | 18 (32.1) | 137 (42.0) |         |
| NHL                             | 5 (8.9)  | 28 (8.6)  |         |
| MDS                             | 6 (10.7) | 36 (11.0) |         |
| Other                           | 19 (33.9) | 60 (19.7)  |         |
| Nonmalignant                    | 1 (1.8)  | 14 (4.3)  |         |
| SAA                             | 1 (1.8)  | 6 (1.8)   |         |
| Other                           | 0 (0.0)  | 8 (2.5)   |         |
| CMV seropositivity, n (%)       | 37 (66.1) | 191 (58.6) | .47     |
| Donor relation, n (%)           | 30 (53.6) | 153 (46.9) | .60     |
| Related                         |        |          |         |
| Cord                            | 4 (7.1)  | 25 (7.7)  |         |
| Unrelated                       | 22 (39.3) | 148 (45.4) | .92     |
| Stem cell source, n (%)         |        |          |         |
| Bone marrow                     | 5 (8.9)  | 38 (11.7) |         |
| Peripheral blood                | 47 (83.9) | 263 (80.7) |         |
| Umbilical cord blood            | 4 (7.1)  | 25 (7.7)  |         |
| Conditioning regimen, n (%)     |        |          |         |
| Radiotherapy-based              | 16 (28.6) | 82 (25.2)  | .71     |
| Chemotherapy only               | 40 (71.4) | 244 (74.8) | .71     |
| Conditioning intensity, n (%)   |        |          |         |
| Myeloablative                   | 28 (50.0) | 161 (49.4) | .52     |
| Nonmyeloablative                | 11 (19.6) | 43 (13.2)  |         |
| Reduced intensity               | 17 (30.4) | 119 (36.5) |         |
| T cell depletion, n (%)         | 22 (39.3) | 140 (42.9) | .71     |
| Acute GVHD, n (%)               | 28 (50.0) | 151 (46.3) | .72     |
| Grade I                         | 13 (46.4) | 64 (42.4)  |         |
| Grade II                        | 10 (35.7) | 54 (35.8)  |         |
| Grade III                       | 4 (14.3)  | 22 (14.6)  |         |
| Grade IV                        | 1 (3.6)   | 11 (7.3)   |         |

ALL indicates acute lymphoblastic leukemia; AML, acute myelogenous leukemia; MDS, myelodysplastic syndrome; NHL, non-Hodgkin lymphoma; SAA, severe aplastic anemia.

* P value calculated using the Wilcoxon rank-sum test.
† P value calculated using the χ² test comparing categories (malignant versus nonmalignant).
Table 2
Clinical Characteristics of vRTI and Outcomes by Viral Pathogen

| Variable                             | RSV   | Influenza A | Influenza B | Rhinovirus |
|--------------------------------------|-------|-------------|-------------|------------|
| Number of episodes (%)               | 11(16.9) | 7(10.8) | 2(3.1) | 45(69.2) |
| Number of patients (%)               | 10(17.9) | 3(5.4) | 2(3.6) | 41(71.2) |
| Initial site of presentation, n/N (%)|       |            |            |            |
| URTI                                 | 8(11[72.7]) | 4(7[57.1]) | 1(2[50.0]) | 25(45[55.6]) |
| LRTI                                 | 3(11[27.3]) | 3(7[42.9]) | 1(2[50.0]) | 18(45[40.0]) |
| Progression to LRTI (from URTI), n/N (%)| 2(25.0)  | 1(425.0)  | 0(0.0)  | 8(2532.0)  |
| Symptoms, n/N (%)                    |       |            |            |            |
| Fever                                | 3(11[27.3]) | 6(7[85.7]) | 2(2[100]) | 15(45[33.3]) |
| Shortness of breath                  | 0(0.0) | 0(0.0) | 0(0.0) | 3(45[6.7]) |
| Cough                                | 10(11[90.9]) | 6(7[85.7]) | 2(2[100]) | 33(45[73.3]) |
| Sputum                               | 2(11[18.2]) | 2(7[28.6]) | 1(2[50.0]) | 14(45[31.1]) |
| Coryza                               | 4(11[36.4]) | 3(7[42.9]) | 0(0.0) | 16(45[35.6]) |
| Antiviral therapy, n/N (%)           | 4(11[36.4]) | 7(7[100]) | 2(2[100]) | 1(45[2.2]) |
| Presence of copathogen, n/N (%)      | 1(11[9.1]) | 7(17[14.3]) | 0(0.0) | 8(45[17.8]) |
| Bacterial                            | 1(11[9.1]) | 7(17[14.3]) | 0(0.0) | 6(45[13.3]) |
| Fungal                               | 0(0.0) | 0(0.0) | 0(0.0) | 6(45[13.3]) |
| Viral                                | 0(0.0) | 0(0.0) | 0(0.0) | 2(45[4.4]) |
| Outcomes, n/N (%)                    |       |            |            |            |
| ICU admission                        | 3(11[27.3]) | 1(7[14.3]) | 0(0.0) | 7(45[15.6]) |
| Use of mechanical ventilation        | 1(11[9.1]) | 1(7[14.3]) | 0(0.0) | 7(45[15.6]) |
| Nonattributable death‡               | 0(0.0) | 0(0.0) | 0(0.0) | 5(41[12.2]) |

* Includes 1 episode in which both RSV and rhinovirus were detected concurrently.
† Percentages do not sum to 100 because 2 episodes of rhinovirus infection were identified as neither URTI nor LRTI, because there was insufficient information available for classification.
‡ Nonattributable death was calculated using the number of patients.

Table 3
Univariate and Multivariate Logistic Regression Analysis of Risk Factors for vRTI

| Variable                           | Category | vRTI, n | Non-vRTI, n | Univariable OR | 95% CI     | P Value | Multivariable OR | 95% CI     | P Value |
|------------------------------------|----------|---------|-------------|----------------|------------|---------|-----------------|------------|---------|
| Age, yr                            | ≤50      | 37      | 184         | 1.00           |            |         |                 |            |         |
|                                    | ≥50      | 19      | 142         | 0.67           | 0.36-1.19  | .18     |                 |            |         |
| Sex                                | Female   | 19      | 141         | 1.00           |            |         |                 |            |         |
|                                    | Male     | 37      | 185         | 1.48           | 0.83-2.74  | .19     |                 |            |         |
| Underlying disease                 | Malignant| 55      | 312         | 1.00           |            |         |                 |            |         |
|                                    | Nonmalignant| 1      | 14          | 0.41           | 0.02-2.08  | .39     |                 |            |         |
| CMV status                         | Negative | 19      | 134         | 1.00           |            |         |                 |            |         |
|                                    | Positive | 37      | 191         | 1.17           | 0.76-2.52  | .30     |                 |            |         |
| Donor relation                     | Related  | 30      | 153         | 1.00           |            |         |                 |            |         |
|                                    | Cord     | 4       | 25          | 0.82           | 0.23-2.29  | .72     | 0.79            | 0.21-2.35 | .69     |
|                                    | Unrelated| 22      | 148         | 0.76           | 0.41-1.37  | .36     | 0.79            | 0.42-1.47 | .46     |
| Stem cell source                   | Bone marrow | 5    | 38          | 1.00           |            |         |                 |            |         |
|                                    | Peripheral blood | 47 | 263 | 1.36 | 0.55-4.10 | .54 |            |            |         |
|                                    | Umbilical cord | 4  | 25         | 1.22           | 0.28-5.03  | .79     |                 |            |         |
| Radiotherapy-based conditioning    | No       | 40      | 244         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 16      | 82          | 1.19           | 0.62-2.20  | .59     |                 |            |         |
| Chemotherapy-only conditioning     | No       | 16      | 82          | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 40      | 244         | 0.84           | 0.45-1.62  | .59     |                 |            |         |
| Conditioning intensity             | Myeloablative | 28   | 161         | 1.00           |            |         |                 |            |         |
|                                    | Non-myeloablative | 11  | 43         | 1.47           | 0.66-3.13  | .33     |                 |            |         |
|                                    | Reduced intensity | 17   | 119        | 0.82           | 0.42-1.56  | .55     |                 |            |         |
| T cell depletion                   | No       | 35      | 186         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 21      | 132         | 0.86           | 0.48-1.53  | .61     |                 |            |         |
| Acute GVHD                         | No       | 28      | 175         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 28      | 151         | 1.16           | 0.66-2.05  | .61     | 1.10            | 0.61-2.00 | .75     |
| Acute GVHD grade                   | 1        | 13      | 64          | 1.00           |            |         |                 |            |         |
|                                    | 2        | 10      | 54          | 0.91           | 0.36-2.24  | .84     |                 |            |         |
|                                    | 3        | 4       | 22          | 0.90           | 0.23-2.84  | .86     |                 |            |         |
|                                    | 4        | 1       | 11          | 0.45           | 0.02-2.62  | .46     |                 |            |         |
| Lymphopenia                        | No       | 29      | 160         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 24      | 151         | 0.88           | 0.49-1.57  | .66     | 0.89            | 0.48-1.62 | .70     |
| Neutropenia                        | No       | 36      | 199         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 17      | 112         | 0.84           | 0.44-1.54  | .58     |                 |            |         |
| Previous autograft                 | No       | 42      | 281         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 14      | 45          | 2.08           | 1.03-4.05  | .04     | 2.14            | 1.05-4.20 | .03     |
| Previous allograft                 | No       | 55      | 319         | 1.00           |            |         |                 |            |         |
|                                    | Yes      | 7       | 1           | 0.83           | 0.04-4.78  | .86     |                 |            |         |

Boldfaced values indicate significance.
CI indicates confidence interval; OR, odds ratio.
mediated by IFN-γ, appear to have a protective role in RSV infections, whereas Th2 responses may be associated with more severe disease manifestations [28]. The profound immune depletion and impaired immune responses seen in the acute period following transplantation could account for the rates of initial LRTI and progression seen even with respiratory viruses considered less pathogenic, such as rhinoviruses. In addition, our cohort had a lower rate of antiviral therapy for RSV. Because we did not evaluate pulmonary function test results, the risk for vRTI and later complications, such as airway disease, as shown previously by Erard et al. [30], could not be assessed.

A limitation of this study was the number of RVs included in the multiplex PCR panel. Other RVs, such as parainfluenza virus, adenovirus, coronavirus, and hMPV, were not routinely tested but might be important causes of morbidity in these patients. As such, rhinovirus as a cause of vRTI and its burden in the study may be overestimated. Furthermore, because in this study we examined only prevalence, risk factors, and outcomes of vRTI within 100 days following transplantation our results might be not translatable beyond this time frame. However, because this is a known high-risk period [23,31], our study focused on this time frame to highlight the importance of early preventive measures and infection control measures among patients as well as healthcare workers following transplantation.

In conclusion, vRTI was prevalent among our allo-HSCT recipients. Although its impact on mortality appears limited, it led to a number of ICU admissions and necessitated the use of mechanical ventilation in several cases. Our findings indicate that an association between previous auto-HSCT and an increased risk of vRTI. Outcomes were poorest for patients diagnosed within 30 days following transplantation, a group of patients at high risk and with a poor prognosis for whom early preventive and infection control measures should be targeted.

ACKNOWLEDGMENTS

The authors thank Ms. Jenny Collins for her assistance with the transplantation-related data and the staff of Melbourne Health Shared Pathology Service for the pathology information. Aspergillus PCR testing was performed by the Department of Microbiology at Westmead Hospital.

Financial disclosure: The World Health Organization Collaborating Centre for Reference and Research on Influenza is supported by the Australian Government Department of

Figure 2. Kaplan-Meier curves showing the probability of survival by vRTI status. The log-rank test was used to assess differences in the survival to death (A) and ICU admission (B) among 382 patients within 100 days after transplantation.
Health, B.W.T. is supported by a Peter MacCallum Cancer Centre Clinical Research Fellowship and a National Health and Medical Research Council Early Career Fellowship.

Conflict of interest statement: There are no conflicts of interest to report.

REFERENCES
1. Chemaly RF, Ghosh S, Bodey GP, et al. Respiratory viral infections in adults with hematologic malignancies and human stem cell transplantation recipients: a retrospective study at a major cancer center. Medicine (Baltimore). 2006;85:278-287.
2. Hassan IA, Chopra R, Sweeney R, Mutton KJ. Respiratory viral infections after bone marrow/Peripheral stem-cell transplantation: the Christie Hospital experience. Bone Marrow Transplant. 2003;32:73-77.
3. Ljungman P, Ward KN, Crooks BN, et al. Respiratory virus infections after stem cell transplantation: a prospective study from the Infectious Diseases Working Party of the European Group for Blood and Marrow Transplantation. Bone Marrow Transplant. 2001;28:479-484.
4. Martino R, Ramírez E, Rabella N, et al. Respiratory virus infections in adults with hematologic malignancies: a prospective study. Clin Infect Dis. 2003;36:1-8.
5. Whimbey E, Champlin RE, Couch RB, et al. Community respiratory virus infections among hospitalized adult bone marrow transplant recipients. Clin Infect Dis. 1996;22:778-782.
6. Champlin RE, Whimbey E. Community respiratory virus infections in bone marrow transplant recipients: the M.D. Anderson Cancer Center experience. Biol Blood Marrow Transplant. 2001;7(Suppl):85-105.
7. Ljungman P. Respiratory virus infections in bone marrow transplant recipients: the European perspective. Am J Med. 1997;102:44-47.
8. Martino R, Porras RP, Rabella N, et al. Prospective study of the incidence, clinical features, and outcome of symptomatic upper and lower respiratory tract infections by respiratory viruses in adult recipients of hematopoietic stem cell transplants for hematologic malignancies. Biol Blood Marrow Transplant. 2005;11:781-796.
9. Kupfers J, Campbell AP, Cest A, Corey L, Boechl M. Comparison of conventional and molecular detection of respiratory viruses in hematopoietic cell transplant recipients. Transpl Infect Dis. 2009;11:298-303.
10. Gunson RN, Collins TC, Carman WF. Real-time RT-PCR detection of 12 respiratory viral infections in four tripleplex reactions. J Clin Virol. 2005;33:341-344.
11. Templeton KE, Scheltinga SA, Beermsa MF, Kroe AC, Claas EC. Rapid and sensitive method using multiplex real-time PCR for diagnosis of infections by influenza A and influenza B viruses, respiratory syncytial virus, and parainfluenza viruses 1, 2, 3, and 4. J Clin Microbiol. 2004;42:1564-1569.
12. White PL, Linton Cj, Perry MD, Johnson EM, Barnes RA. The evolution and evaluation of a whole blood polymerase chain reaction assay for the detection of invasive aspergillosis in hematology patients in a routine clinical setting. Clin Infect Dis. 2006;42:479-486.
13. Centre for International Blood and Marrow Transplant Research. Form Instruction Manual. Vol 2017. Available at: https://www.cibmtr.org/manuals/fim?en=1s=1i=en-. Accessed June 13, 2017.
14. Hirsch HH, Martino R, Ward KN, Boechl M, Einsele H, Ljungman P. Fourth European Conference on Infections in Leukaemia (ECIL-4): guidelines for diagnosis and treatment of human respiratory syncytial virus, parainfluenza virus, metapneumovirus, rhinovirus, and coronavirus. Clin Infect Dis. 2013;56:258-266.
15. Teh BW, Worth LJ, Harrison SJ, Thursky KA, Slavin MA. Risks and burden of viral respiratory tract infections in patients with multiple myeloma in the era of immunomodulatory drugs and bortezomib: experience at an Australian Cancer Hospital. Support Care Cancer. 2015;23:1901-1906.
16. Ljungman P, de la Camara R, Perez-Bercoff I, et al. Outcome of pandemic H1N1 infections in hematopoietic stem cell transplant recipients. Haematologica. 2011;96:1231-1235.
17. Wolfrom AM, Porcher R, Legoff J, et al. Viral respiratory infections diagnosed by multiplex PCR after allogeneic hematopoietic stem cell transplantation: long-term incidence and outcome. Biол Blood Marrow Transplant. 2014;20:1238-1241.
18. Ison MG, Hayden FG, Kaiser L, Corey L, Boechl M. Rhinovirus infections in hematopoietic stem cell transplant recipients with pneumonia. Clin Infect Dis. 2003;36:1139-1143.
19. Griffiths C, Drews SJ, Marchant DJ. Respiratory syncytial virus: infection, detection, and new options for prevention and treatment. Clin Microbiol Rev. 2017;30:277-319.
20. Gorschützer M, Glasmacher A, Sarazin S, et al. CD4+ T lymphocyte counts after autologous transplantation in multiple myeloma: a retrospective study. Leuk Lymphoma. 2007;48:506-512.
21. Schütz P, Brandhorst D, Stellberg W, et al. Immune parameters in multiple myeloma patients: influence of treatment and correlation with opportunistic infections. Leuk Lymphoma. 2006;47:1570-1582.
22. Bredius RG, Templeton KE, Scheltinga SA, Claas EC, Kroe AC, Vossen JM. Prospective study of respiratory viral infections in pediatric hematopoietic stem cell transplantation patients. Pediatr Infect Dis J. 2004;23:518-522.
23. Chakrabarti A, Avivi I, Mackinnon S, et al. Respiratory virus infections in transplant recipients after reduced-intensity conditioning with Campath-1H: high incidence but low mortality. Br J Haematol. 2002;112:1125-1132.
24. Srinivasan A, Wang C, Yang J, Shene JL, Leung WH, Hayden RT. Symptomatic parainfluenza virus infections in children undergoing hematopoietic stem cell transplantation. Biol Blood Marrow Transplant. 2011;17:1520-1527.
25. Protheroe RE, Kirkland KE, Pearce RM, et al. The clinical features and outcome of 2009 H1N1 influenza infection in allo-SCT patients: a British Society of Blood and Marrow Transplantation Study. Bone Marrow Transplant. 2012;47:88-94.
26. Khanna N, Steffen I, Steid JD, et al. Outcome of influenza infections in outpatients after allogeneic hematopoietic stem cell transplantation. Transpl Infect Dis. 2009;11:100-105.
27. Rihani R, Hayahane W, Sultan I, et al. Infections with the 2009 H1N1 influenza virus among hematopoietic SCT recipients: a single center experience. Bone Marrow Transplant. 2011;46:1430-1436.
28. Avetisyan G, Mattsson J, Sparrelid E, Ljungman P. Respiratory syncytial virus infection in recipients of allogeneic stem-cell transplantation: a retrospective study of the incidence, clinical features, and outcome. Transplantation. 2009;88:1222-1226.
29. Russell CD, Unger SA, Walton M, Schwarze J. The human immune response to respiratory syncytial virus infection. Clin Microbiol Rev. 2017;30:481-502.
30. Erard V, Chien JW, Kim HW, et al. Airflow decline after myeloablative allogeneic hematopoietic cell transplantation: the role of community respiratory viruses. J Infect Dis. 2006;193:1619-1625.
31. Ghosh S, Champlin R, Couch R, et al. Rhinovirus infections in myelosuppressed adult blood and marrow transplant recipients. Clin Infect Dis. 1999;29:528-532.