Respiratory Virus Infections in Pediatric Hematopoietic Stem Cell Transplantation

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Respiratory virus infections (RVI) have become an increasingly appreciated problem in the hematopoietic stem cell transplant (HSCT) population. A retrospective analysis of 274 patients undergoing 281 HSCT at St. Jude Children’s Research Hospital from January 1994 through December 1997 was performed. Medical and clinical laboratory records were reviewed beginning at the onset of conditioning through the year following each HSCT, and the analysis was done for the first RVI only. Thirty-two (11%) of 281 HSCT cases developed a RVI during the first year post-HSCT. The most frequent cause of RVI was human parainfluenza virus type 3. Univariate analysis was performed to determine the association between risk factors and the cumulative incidence of RVI. Respiratory viruses are frequent causes of infections in the first year post-HSCT in the pediatric population. Only allogeneic transplant and the degree of acute or chronic graft versus host disease were found to be statistically significant risk factors for RVI.

Respiratory infections are a significant source of morbidity and mortality in the hematopoietic stem cell transplant (HSCT) recipient [1, 2]. Respiratory infections in HSCT patients have been attributed to bacteria, fungi and other opportunistic pathogens [3, 4]. Regarding viruses, the focus has been predominantly on cytomegalovirus (CMV) and, to a lesser extent, on the adenoviruses [5–7].

Community respiratory viruses such as respiratory syncytial virus (RSV), influenza, human parainfluenza (hPIV), and picornaviruses are recognized as important causes of respiratory disease in the adult HSCT recipient [8–17]. Distinctive features of respiratory viral infections (RVIs) among immunocompromised patients are a high frequency of nosocomial acquisition, prolonged persistence of infection, high frequency of progression to pneumonia, and a high mortality rate [11].

There is a paucity of information regarding RVIs in the pediatric HSCT population [18, 19]. To assess the burden of disease caused by RVIs in pediatric HSCT recipients, we retrospectively evaluated this patient group at St. Jude Children’s Research Hospital (SJCRH) for 4 consecutive years. The present study describes the cumulative incidence, clinical characteristics, risk factors, and outcome of RVIs during the first year after HSCT in pediatric patients.

METHODS

Patients. The medical and virology laboratory records of consecutive patients undergoing HSCT at SJCRH from 1 January 1994 through 31 December 1997...
were retrospectively analyzed to identify patients with positive respiratory virus cultures. The records were reviewed for the conditioning period and a year following each HSCT. During the study period, 290 consecutive HSCTs were performed on 274 patients at SJCRH. Sixteen patients required a second HSCT for loss of graft or disease recurrence. The second transplant was included in the study only if it occurred >1 year after the initial HSCT. Seven such patients were identified, resulting in a total of 281 HSCT recipients evaluated.

Patient demographics, primary disease, date, and type of HSCT, clinical characteristics, type and season of the RVI, antiviral therapy, and patient outcome were recorded. We also investigated whether the RVIs had been hospital or community acquired. The only viruses evaluated in the study were adenovirus, hPIV, influenza, and RSV, because during the study period rhinoviruses or nonpolio enteroviruses were not routinely cultured. All respiratory viral isolates were associated with respiratory symptoms, except for 3 cases of asymptomatic adenoviral infections that were not included in the RVI group.

Methods of transplantation. All the patients underwent antineoplastic and immunosuppressive conditioning as appropriate for their primary condition and indication for HSCT, as described elsewhere [7]. Recipients of allogeneic HSCT received prophylaxis and treatment against graft versus host disease (GVHD) as described elsewhere [7]. Mismatched family member (MFM) and matched unrelated donor (MUD) transplants were T-cell depleted. Patients were placed in individual rooms with high-efficiency particulate air filtration. Strict hand-washing precautions were followed. Empiric systemic antibacterial and antifungal agents were used routinely. Ganciclovir was given for prophylaxis against CMV beginning after engraftment and continuing until day +120 for patients who were CMV positive or received a transplant from a CMV-positive donor. Prophylactic intravenous immunoglobulin (IVIG) was used routinely in allogeneic transplant recipients, weekly from day of transplant until 120 days post-HSCT and then monthly up to 1 year posttransplant for MUD and MFM recipients.

Definitions. “Respiratory viral infection” was defined as the laboratory identification of a respiratory pathogen in association with new onset or exacerbation of respiratory symptoms. Upper respiratory tract illness (URI) was defined as the presence of coryza, pharyngitis, sinusitis, and/or cough with a clear chest radiograph. Lower respiratory tract illness (LRI) was defined as the presence of stridor, wheezing, hypoxia, or pneumonia with a new radiographic infiltrate. Infections were considered to be nosocomial when they developed ≥48 h after hospital admission. Adenoviral infections were defined according to established criteria and were not divided into community or hospital acquired, because this virus can reactivated genodously [6]. Acute and chronic GVHD were defined and graded according to published criteria. Mild acute GVHD was described as grades 0 or 1, moderate as grade 2, and severe acute GVHD as grades 3 or 4 [20, 21].

Neutropenia was defined as an absolute count of <500 neutrophils and band forms/μL of blood. Engraftment was defined as the presence of >500 neutrophils and band forms/μL of blood on 2 consecutive days.

Virological techniques. Surveillance viral cultures were obtained twice weekly during hospitalization from the throat, nasopharynx, and blood. These cultures, as well as tracheal, bronchoalveolar lavage (BAL), and pleural fluid culture specimens, were also obtained when clinically indicated. Lung biopsy and autopsy specimens were routinely sent for viral culture. Nasopharyngeal swabs for culture were obtained when clinically indicated in outpatients. Patients with positive cultures had follow-up cultures obtained until 2 consecutive specimens had no viral growth.

Fluorescent antibody (FA) and viral culture was performed on all the nasopharyngeal swab samples. Specimens for FA staining were collected with a flexible shaft Dacron swab and transported in 0.5 mL of sterile normal saline. Specimens for viral culture from respiratory tract sites were placed into 3 mL of viral transport media. Samples were transported to the laboratory on ice and inoculated within 4 h onto tissue cultures (human lung carcinoma cells [A549], human laryngeal epidermoid carcinoma cells [HEp-2], human embryonic lung tissue cells [MRC5], and rhesus monkey kidney cells [RMK]). Cell lines were observed for cytopathic effect for 21 days. Shell vial assays were performed to enhance the time to detection for hPIV, influenza, and RSV. Hemadsorption (HA) staining was done at 7 and 14 days of culture. FA was also performed to confirm positive cultures or HA staining.

Statistical analysis. For the analysis, 281 HSCT cases were identified. Seven patients had 2 HSCT >1 year apart and were analyzed as independent units for the incidence of RVIs. We assessed the cumulative incidence (CI) of any type of first RVI. The time during which a patient was at risk of first RVIs was defined as the time from the date of HSCT to the date of any type of RVI, the date of death or the date of 1 year follow-up, whichever came first. Death due to any cause was considered to be a competing risk. For the analysis of specific type of the first RVI, once an RVI was identified (event), subsequent cases with other types of RVIs were excluded. The methods described by Pentice and Gray were used to estimate the CI of RVIs and to compare the CI of RVIs between the risk factors, respectively [22, 23]. The risk factors we assessed were age group, sex, type of HSCT, season of the HSCT, and severity (no or grades 0–2 vs. grades 3 and 4) of GVHD. For the age-group evaluation, patients were distributed as follows: 0–5, 5–15 and >15 years of age. The seasons were divided by groups of 3 calendar
months, with winter including January through March and so on for the other seasons.

RESULTS

The characteristics of the patients included in the study are listed in table 1. There was a slight predominance of males, and the racial distribution reflects the population treated at SJCRH. Patients underwent HSCT for a variety of diseases that included hematologic malignancies, solid tumors as well as other conditions including sickle cell disease, metabolic disorders, and primary immunodeficiencies (table 1).

During the first year post-HSCT, 32 episodes of RVI were diagnosed. Four patients had 2 nonsimultaneous RVIs within the first year post-HSCT; however, only the first episode was included in the statistical analysis. These 4 episodes were included in the total number of isolates and the description of clinical characteristics and outcome (table 2). The hPIV were the most frequently isolated viruses (47%), followed by adenovirus (19%), influenza A (17%), and RSV (14%; table 2).

RVIs occurred throughout the year and were not specifically confined to the winter months (figure 1). Respiratory viruses were generally isolated during the season in which they normally occur in healthy pediatric hosts. Influenza viruses were isolated in the winter, hPIV-3 predominated during the spring and summer, the other parainfluenza viruses in the fall, and RSV tended to occur in early winter. In contrast, adenovirus occurred year round.

**Clinical characteristics.** All the patients, except for 2 cases of adenoviral illness, presented with URI symptoms, with 10 (28%) of the episodes progressing to pneumonia. Nine of the infections were acquired nosocomially (table 2). Of the 20 patients who acquired the infection in the community after hospital discharge, half of them were readmitted to the hospital.

Of the 7 patients who had 2 HSCTs >1 year apart, 4 developed an RVI during the course of the second transplant. Three of them had an autologous HSCT the first time and a MUD-HSCT the second time; the other 1 had a matched sibling HSCT on both occasions.

Most of the cases (22 [61%] of 36 cases) had a single positive culture. In the cases with multiple positive cultures, viral shedding ranged from 2 to 38 days. The majority of patients had positive cultures from 1 site, with a maximum of 4. Most viruses were isolated from nasopharyngeal specimens.

**Parainfluenza viruses.** There were 12 cases of hPIV-3 infection, with 8 patients presenting exclusively with URI symptoms. The 3 patients who had a hospital-acquired infection progressed to pneumonia, and 2 of them had prolonged shedding of the virus, 1 for 33 days and another for 38 days, with persistence of respiratory symptoms. Only 1 patient presented with croup.

Two patients required admission to the intensive care unit (ICU), 1 of them with pneumonia and the 1 with croup. Two of the patients with pneumonia received ribavirin therapy. One of them required ventilator support and died from hPIV-3 pneumonitis, with the virus being isolated from BAL, lung biopsy, and autopsy specimens.

Two patients had infection with hPIV-1, both hospital-acquired, during the neutropenic phase at 16 days and 17 days posttransplant, respectively. Both started with URI symptoms and progressed to pneumonia. Only 1 received ribavirin therapy, and both survived the infection.

There were 3 cases of hPIV-4 infection, all in allogeneic HSCT patients. All of them were hospital acquired, between days 8 and 17 post-HSCT. All presented with URI symptoms, with 1 of them progressing to pneumonia and 1 developing...
Table 2. Clinical characteristics and outcomes for 36 episodes of respiratory viral infection (RVI) during first year post–hematopoietic stem cell transplantation (HSCT).

<table>
<thead>
<tr>
<th>Finding</th>
<th>Type of virus</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>hPIV-1</td>
</tr>
<tr>
<td>No. of cases (%)</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Age, years</td>
<td></td>
</tr>
<tr>
<td>0–5</td>
<td>1</td>
</tr>
<tr>
<td>5–15</td>
<td>0</td>
</tr>
<tr>
<td>&gt;15</td>
<td>1</td>
</tr>
<tr>
<td>Source of RVI</td>
<td></td>
</tr>
<tr>
<td>Community</td>
<td>0</td>
</tr>
<tr>
<td>Nosocomial</td>
<td>2</td>
</tr>
<tr>
<td>Graft when RVI</td>
<td></td>
</tr>
<tr>
<td>Pre-engraftment</td>
<td>2</td>
</tr>
<tr>
<td>Engraftment</td>
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</tr>
<tr>
<td>Type of HSCT</td>
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</tr>
<tr>
<td>Autologous</td>
<td>0</td>
</tr>
<tr>
<td>Matched sibling</td>
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</tr>
<tr>
<td>MMFM</td>
<td>0</td>
</tr>
<tr>
<td>MUD</td>
<td>2</td>
</tr>
<tr>
<td>Days after HSCT</td>
<td></td>
</tr>
<tr>
<td>&lt;30</td>
<td>2</td>
</tr>
<tr>
<td>30–100</td>
<td>0</td>
</tr>
<tr>
<td>&gt;100</td>
<td>0</td>
</tr>
<tr>
<td>Outcome</td>
<td></td>
</tr>
<tr>
<td>Pneumonia (%)</td>
<td>2 (100)</td>
</tr>
<tr>
<td>Death (%)</td>
<td>0</td>
</tr>
</tbody>
</table>

NOTE. Data shown are no. of episodes. Flu-A, influenza A virus; Flu-B, influenza B virus; hPIV, human parainfluenza virus; MMFM, mismatched family member; MUD, matched unrelated donor; NA, not applicable; RSV, respiratory syncytial virus.

both pneumonia and croup. Although 1 patient required mechanical ventilation, none received ribavirin therapy and all survived the infection.

Adenovirus. In this series, there were 7 adenoviral infections. Four patients had only URI symptoms, and adenovirus was exclusively isolated from samples of the respiratory tract. Two patients had evidence of disseminated adenoviral disease with lower respiratory tract involvement requiring ICU support, and 1 had exclusively LRI. One of the patients with disseminated disease received ribavirin but died despite therapy.

Influenza. Six patients were infected with influenza A—in all cases the infection was community acquired—between days 114 and 315 post-HSCT. Five patients were readmitted to the hospital, and 4 were treated with either amantidine or rimantidine. None of the patients had clinical or radiographic evidence of LRI.

The only patient with influenza B infection acquired the infection nosocomially presenting at day 1 posttransplant and rapidly progressing to pneumonia and respiratory failure. Despite intensive supportive therapy and compassionate treatment with zanamivir the patient expired of complications of influenza B pneumonitis [24].

Respiratory syncytial virus. Five patients were infected with RSV; all acquired the infection as outpatients between days 42 and 335 post–HSCT. Three of those patients were readmitted to the hospital to receive aerosolized ribavirin therapy. All RSV-infected patients had exclusively URI symptoms, with normal chest radiographs, and none of them progressed to pneumonia. Four were diagnosed by fluorescent assay performed on the original specimen.

The acquisition of RVI seemed to be evenly distributed among the 3 described phases of immunologic recovery post–HSCT. One-third of the infections occurred during the neutropenic phase (days 0–30), the early engraftment phase (days 31–100), and the late engraftment phase (days 101–365), respectively. The risk for development of LRI appeared to be higher, however, if the RVI was acquired prior to engraftment. Of the 11 patients who developed an RVI pre-engraftment, 7 (64%) progressed to LRI, compared to 5 of 25 patients (20%) developing LRI if their RVI was acquired after engraftment.
In addition, allogeneic transplant recipients having a 5-fold higher CI than autologous transplant patients with occurrence of RVI, with allogeneic transplant recipients having a 5-fold higher CI than autologous transplant patients with occurrence of RVI. The CI for the development of the first RVI at 1 year posttransplant was 11.7 ± 2% (table 2).

Risk factors. Univariate analysis was performed to determine the risk factors for RVI after HSCT. There was no statistically significant difference on the CI of the first RVI among sex (P = .2), age group (P = .99), and season of the HSCT (P = .26); however, the type of transplant highly correlated with occurrence of RVI, with allogeneic transplant recipients having a 5-fold higher CI than autologous transplant patients (P < .001; figure 1). In addition, allogeneic transplant recipients with grade 3–4 GVHD also had a significantly higher CI of first RVI than allogeneic transplant recipients with no or grades 1 or 2 GVHD (P = .02).

Mortality. Ten of the patients with RVIs died within the first year after the transplant, for a case-fatality ratio (CFR) of 31.3%. That CFR is quite similar to the observed CFR in the transplant population as a whole during the same sample period (75 [27.4%] of 274 patients). Of these 10 deaths, only 3 were directly attributable to an RVI, 1 case each of influenza B, hPIV-3, and adenovirus. Thus, the overall CFR attributable to RVI was 3 (1.1%) of 274. Mortality from RVI tended to occur with infections acquired early in the transplant period (range, day −1 to day +37).

DISCUSSION

As our analyses demonstrate, RVIs are a frequent complication in children undergoing HSCT. Eleven percent of our HSCT cases developed an RVI in the first year posttransplant. This figure may underestimate the real incidence of RVIs, because such errors are inherent to retrospective studies. The results of our study suggest that respiratory viruses are important etiologies of potentially serious acute respiratory illnesses in pediatric HSCT patients. During the study period there were also 3 cases of CMV pneumonitis and 1 case of varicella-zoster virus pneumonitis, demonstrating the importance of respiratory viruses as a source of morbidity in HSCT patients when ganciclovir prophylaxis is used routinely.

An interesting finding of our study was the high percentage of hPIV isolates, especially type 3, from patients with RVIs. This is higher than has been reported in previous pediatric series of HSCT patients [18, 19] but similar to what has been reported in some adult series [8, 15]. Although our study exclusively examined children, our incidence of RSV was low when compared to adult series where RSV has been the predominant virus [9].

An explanation may involve the fact that we did not have any nosocomial influenza and RSV infections compared to adult studies where significant outbreaks with these viruses have been described. Published data from series of HSCT in adults indicate that 55%–74% of all RSV and influenza A infections were nosocomial [13, 14, 25]. We found hPIV to be the predominant nosocomial pathogen, although we could not identify an outbreak during the study period.

Other explanation for the low incidence of RVIs in this population is the possibility that in mild URI cases the patients did not seek medical attention. Another factor could be the routine use of IVIG during the study period that might have prevented some cases of RVIs.

Age was not a risk factor for acquisition of RVIs (table 1). There was no predominance of younger children as is seen in the normal pediatric population. The seasonal nature of RVIs appears to extend to the HSCT population. One of our hypotheses was to establish whether patients undergoing HSCT during the winter months had a higher incidence of RVI. That did not seem to be the case, the incidence of RVIs being similar among the different seasons. The whole year should be considered high-risk for RVIs in pediatric HSCT recipients, with some viruses like RSV and the influenza viruses predominating in the winter, the hPIV doing so in late spring, summer, and fall, and adenoviruses occurring throughout the year.

The only risk factors for RVIs found to be statistically significant were being the recipient of an allogeneic transplant and developing severe GVHD (grade 3 or 4). This is probably related to the higher degree of immunosuppression that these patients have secondary to their conditioning regimens and the treat-
ment of GVHD. It may also be related to longer hospital stays and hence greater frequency of viral culture in the patients with GVHD. These risk factors have also been identified in smaller studies of pediatric HSCT recipients [7, 19].

The morbidity and mortality associated with RVIs were substantial. More than a quarter (28%) of these infections were complicated by pneumonia and ~5% were complicated by croup. The progression to pneumonia when the RVI occurred within the first 30 days posttransplant (64%) was similar to what has been reported in adults (58%–70%), but the outcome seems to be better in these patients when compared to adults [9, 14–16]. The lack of specific CI rates of RVI reported in adult HSCT recipients does not allow direct comparison between our series and adult populations.

The mortality of patients acquiring an RVI in our study was 9.4% (3 out of 32 patients), which is similar to the CFR (14%) reported in a recent series of pediatric HSCT patients [19]. Pediatric solid organ transplant recipients have also been reported to have similar CFR (0%–19%) for community-acquired RVIs [26–29]. Comparisons with this population should be undertaken with caution, however, because of likely differences in surveillance techniques. The CFR in our study, however, is lower than the CFR (22%–36%) reported in published series of adult populations [9, 14–16]. Two of the deaths in our study occurred in patients acquiring the infection very early in the transplant period. We would agree with previous recommendations of delaying the transplant, when feasible, until signs and symptoms of URI have resolved [9].

This series of pediatric HSCT recipients with RVIs underscores the need for prevention of RVIs, especially early in the transplant period. Preventive measures such as strict isolation and influenza immunization of staff and families should be observed. Patients should be screened for respiratory symptoms, and viral cultures should be performed as soon as symptoms develop. Patients, staff, and family members with new onset of respiratory symptoms should be isolated to prevent nosocomial transmission to other immunocompromised individuals.

In summary, respiratory viruses are a frequent cause of infection in the first year post-HSCT in pediatric patients, with a CI of 3.9%, 7.2%, and 11.7% at 30, 100, and 365 days post-transplant, respectively. RVIs occur in every season in HSCT recipients, with seasonal variations in the etiology similar to those observed in the general pediatric population. Risk factors for acquiring an RVI after HSCT include being the recipient of an allogeneic transplant and the presence of grades 3–4 acute or chronic GVHD.

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