Antiviral therapy of respiratory viruses in haematopoietic stem cell transplant recipients

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Introduction

The long-term survival of haematopoietic stem cell transplant (HSCT) recipients has significantly increased over the last decade [1]. Outcomes related to respiratory virus infections have also improved, even if significant morbidity and mortality is still described. There are preventive and therapeutic strategies available to reduce morbidity and mortality for influenza, respiratory syncytial virus (RSV) and human adenovirus (HAdV). Parainfluenza virus (PIV) and human metapneumovirus (HMPV) also have treatments, although the benefits of therapy are not clear. The most recent epidemiological studies have shown that human rhinovirus (HRV) and human coronavirus (HCoV) were the most frequently encountered respiratory viruses in HSCT recipients; no drugs are available to treat those two groups of viruses. Similarly, newly discovered human bocavirus (HBoV) and polyomaviruses KI and WU have been found from various specimen types but no clear symptomatic diseases have been associated with these viruses and no treatments have been evaluated.

Diagnosis

Respiratory viral diseases have been traditionally diagnosed by viral culture and direct immunofluorescence testing of respiratory secretions. However, molecular assays targeting viral RNA or DNA have better sensitivity and are now widely available [2]. Advances in the development of diagnostic molecular virology testing have led to recent FDA approval of multiplex PCR assays for respiratory virus detection [3]. Many other multiplex panels and microarrays are being developed and are on the market as analyte-specific reagents [4,5]. Comparative studies among various multiplex panels have shown similar results with high sensitivity compared with viral culture [6,7]. However, comparison with real-time PCR has shown slightly lower sensitivity of multiplex panels with results varying depending on specific viruses [8–11]. Many HSCT recipients shed low quantities of virus for weeks to months, which could potentially be missed with those assays. The clinical significance of these low viral loads has yet to be determined. In one study, detection of viral RNA from serum occurred in 6 out of 66 patients with proven viral pneumonia. These patients had higher viral load in bronchoalveolar lavage samples and an increased risk of death (adjusted rate ratio 1.8; P=0.02) [12]. With the development of new drugs, cell culture will continue to play an important role in diagnosing the emergence of resistance. Testing by antigen detection, such as direct immunofluorescence, in addition to molecular assays, can potentially provide more rapid results, allowing patients to receive a specific drug faster. Optimal
approaches for diagnostic testing to provide rapid and reliable results continue to require institutional assessment of costs, complexity and potential clinical benefit, as well as continued research.

**Epidemiology**

The changing epidemiology of respiratory viral infections in HSCT recipients has been reviewed recently [13]. Most respiratory virus infections occur in 1 to 10% of HSCT recipients during the first 100 days post-transplantation, with the exception of HRV, HCoV and HAdV, with an incidence ranging from 10 to 30% [13]. Patients infected with influenza virus, RSV, PIV and HMPV develop lower respiratory tract infection (LRTI) in 10 to 50% of cases. Patients infected with HAdV have more consistently high rates of pneumonia (30–40%) and patients infected with HRV or HCoV have lower rates (5%). The overall incidence of pandemic H1N1 has not been well-described in HSCT recipients but LRTI was described in approximately 30% of cumulative cases in published studies [13]. Death associated with LRTI caused by respiratory viruses ranges between 10 and 50% depending on the specific virus (Table 1); however, HAdV seems to have the highest related mortality. Severe and lethal cases of HRV and HCoV have been described but are rare [14,15]. Respiratory viral infection early after transplantation, graft-versus-host disease (GVHD), allogeneic HSCT, myeloablative regimen and transplant from a mismatched unrelated donor, all affect the severity of lymphopenia which seems to be the predominant factor in increasing the risk of progression to LRTI and death [16–21]. One large prospective study performing respiratory virus detection by real-time PCR prior to allogeneic HSCT has shown that respiratory viruses are common (25% of patients) and associated with increased mortality, prolonged hospitalization and increased pulmonary morbidity [22].

**Prevention of respiratory viral infection**

Vaccination is generally the method of choice to prevent infections, but licensed vaccines are only available against influenza virus. Immunogenicity of inactivated influenza vaccine is 10–50% lower in patients receiving chemotherapy than healthy adults and response rates in HSCT recipients are as low as 9–31% depending on specific subtypes [23–25]. Patients immediately preceding transplant or in the 6 months following the transplantation are unlikely to respond to influenza vaccination [23]. Adherence to vaccination has also been an issue [26]. Influenza vaccination has to be emphasized among immunocompromised patients and their families as the best and preferred way to prevent influenza infection. Patient and family education improve adherence to vaccine and general infection control [27]. In addition to vaccination, chemoprophylaxis may be used to

<table>
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<th>Effective antiviral treatment available</th>
<th>Available drugs</th>
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<tr>
<td>Influenza</td>
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<td>7–35</td>
<td>15–28</td>
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<td>Osmeltamivir, Zanamivir, Amantadine, Ribavirin, IVIG</td>
<td>IV permanivir, IV zanamivir, DAS181, Favipiravir, Lamivudinir</td>
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<td>7–33</td>
<td>±</td>
<td>Ribavirin, Palivizumab, RSV-IVIG</td>
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<td>Ribavirin, IVIG</td>
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<td>±</td>
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*Table 1. Effect of and therapy for respiratory viruses in HSCT recipients

a Taken from [13]. Ag, antigen detection test; DFA, direct immunofluorescence; IV, intravenous; IVIG, pooled intravenous immunoglobulins; LRTI, lower respiratory tract infection; NA, not applicable; PCR, polymerase cycling reaction; +, effective based on randomized studies in non-haematopoietic stem cell transplant (HSCT) population and retrospective cohort studies in HSCT population; –, not effective; ±, effective based on retrospective cohort studies in HSCT population; ?, absence of significant studies or conflicting results.
prevent influenza infection. The neuraminidase inhibitors oseltamivir and zanamivir are both approved for prophylaxis and can be used in unvaccinated immunocompromised patients exposed to influenza virus A or B. Oseltamivir prophylaxis is well-tolerated in HSCT patients and effectiveness has been shown in healthy adults [28,29]. One randomized, double-blind, placebo-controlled study in HSCT and solid organ transplant recipients receiving oseltamivir 75 mg once a day versus placebo confirmed the effectiveness of prophylaxis in immunocompromised patients by describing a 74.9–88.8% reduction in frequency of laboratory-confirmed infection during the 12-week period of peak influenza circulation in the community [30]. Zanamivir prophylaxis is also effective in healthy adults but no prospective studies are available in HSCT patients [31,32]. Many authorities have recommended influenza chemoprophylaxis in immunocompromised patients during community or nosocomial outbreak [33–35]. Oseltamivir prophylaxis during the 2009 pandemic has been shown to select occasionally resistant strains and therefore should be considered cautiously [36–38].

Prevention of RSV infection using passive immunoprophylaxis has only been studied in children and is not routine in HSCT recipients. One study investigated RSV-specific intravenous immunoglobulin (RSV-IVIG) prophylaxis in high-risk adult HSCT recipients but was not able to determine efficacy because the study was underpowered [39]. Palivizumab prophylaxis is licensed for use in children less than 2 years old, and not specifically indicated for those undergoing HSCT. Immunoprophylaxis with palivizumab (15 mg/kg/dose monthly during RSV season) in young children undergoing HSCT was suggested in 2009 international HSCT guidelines but the cost related to this intervention could limit its application (approximately USD 2,300 for each 100 mg vial) [40]. Use of pooled IVIG to reduce respiratory viral infections has not been specifically investigated. However, the use of IVIG for prevention of bacterial or cytomegalovirus infections has not shown a reduction of all-cause mortality and IVIG administered early after HSCT has been associated with veno-occlusive disease, which would preclude its use in the most critical period for respiratory virus infections [41,42]. The 2009 international HSCT guidelines recommend IVIG for patients with severe hypogammaglobulinaemia (serum immunoglobulin G level <400 mg/dl) but its effect on respiratory virus prevention is unknown [34]. The use of palivizumab for outbreak control has been successfully reported in one study in addition to extended infection control measures [43].

Immunoprophylaxis and chemoprophylaxis for respiratory viruses require further investigation. Strict infection control policies should be implemented and enforced to diminish the burden of respiratory viruses in HSCT recipients. The role of asymptomatic shedding detected by molecular assays during nosocomial outbreaks remains undetermined.

Treatment of respiratory virus infections

Treatment of respiratory viruses in HSCT recipients is extremely challenging. Few drugs are available, clinical trials are difficult to conduct and efficacy data are limited. Nevertheless, new drugs are coming and more information is now available. In high risk patients, preemptive treatment at the stage of upper respiratory tract infection (URTI) to limit progression to LRTI is a common strategy used mainly for influenza virus, RSV and HAdV. However, all respiratory viruses may present with LRTI and treatment could potentially be lifesaving. Whether treatment of asymptomatic shedding may reduce transmission or progression to symptomatic disease is undetermined, and the effect of treatment on risk of long-term complications is not clear (Figure 1). Use of quantitative molecular assays might be useful in the follow-up of infected patients. Determination of viral loads after initiation of therapy can potentially evaluate the response to antiviral therapy and suggest the emergence of resistance (Figure 2). Currently available treatments and new drugs for specific respiratory virus infections are discussed below.

Influenza

Influenza treatment in HSCT recipients has relied on neuraminidase inhibitors (oseltamivir and zanamivir) since adamantane resistance became widespread in 2005–2006 [44,45]. However, seasonal H1N1 circulating between 2007 and 2009 was susceptible to M2-inhibitors (rimantadine and amantadine) and resistant to oseltamivir. Recommendations then suggested either combined therapy with adamantane and oseltamivir or zanamivir monotherapy if influenza subtype was unknown [46]. Later in 2009, oseltamivir-susceptible pandemic influenza A(H1N1) emerged and eventually replaced oseltamivir-resistant seasonal H1N1. That same pandemic strain was also shown to be uniformly resistant to adamantanes. Based on studies showing a low rate of influenza pneumonia (0–5%) when oseltamivir was started early, oseltamivir was recommended again as first-line therapy to treat pandemic H1N1, seasonal H3N2 or influenza B infections [16,47–49]. A recent retrospective study showed that both early (<48 h) and delayed (>48 h) administration of influenza antiviral therapy decreased rates of LRTI, although earlier initiation was more effective [50]. Since this study was retrospective, no specific antiviral regimens were used. Inhaled zanamivir was also
recommended as an equivalent alternative but difficulties in obtaining the drug as well as adverse events in patients with chronic lung disease have limited its use in this patient population [51,52].

During the 2009 pandemic, many HSCT centres experienced a higher incidence than usual of influenza infections because of the absence of vaccine, as well as ongoing community and nosocomial outbreaks. Cases of severe disease were rapidly described and issues concerning dose and duration of treatment had to be faced. The usual dose of 75 mg twice daily was shown to be well absorbed in intensive care unit patients but the impact of GVHD and mucositis on oseltamivir absorption has not been well studied [53]. Doses of 150 mg twice daily have not shown any advantage over standard doses and were associated with slightly more adverse gastrointestinal events in immunocompetent outpatient adults with uncomplicated illness [54,55]. However, HSCT recipients have higher viral loads, longer shedding and possibly reduced absorption, which could theoretically support use of higher doses of oseltamivir. Duration of therapy is usually 5 days in immunocompetent patients, but immunocompromised patients shed viruses for much longer and the risk to progress to pneumonia is still present even after 5 days.

Figure 1. General concepts of respiratory virus treatment in haematopoietic stem cell transplant recipients

Figure 2. Role of diagnostic methods in management of influenza infection in haematopoietic stem cell transplant recipients

An example of a patient developing oseltamivir resistance is shown [37]. DFA, direct immunofluorescence; Inh, inhaled; IV/intravenous; OTV, oseltamivir; RBV, ribavirin; RMT, rimantadine; ZNV, zanamivir.
Relapse of infection after arrest of antiviral therapy is also of concern. Therefore, a course of 10 days therapy or until the patient stops shedding has been suggested.

Adjuvant therapy
The effect of steroids on influenza infection remains unclear. Corticosteroids have been associated with prolonged influenza shedding, although one study of seasonal influenza in HSCT patients showed a protective effect toward progression to pneumonia [16]. Two recent studies from the same group have shown a similar or perhaps lower risk of progression to LRTI in patients receiving high dose steroids [21,50]. Steroids could potentially be beneficial because of anti-inflammatory effects in patients with acute lung injury or acute respiratory distress syndrome. Another group has shown increased mortality and infection associated with steroid therapy in critically ill patients but the study was not limited to immunocompromised patients and these data are difficult to apply to the HSCT population [56]. Data from randomized trials would be very helpful to clarify the effect of steroids in HSCT patients infected with influenza. Similarly, no randomized trials have demonstrated benefits from IVIG as adjuvant treatment but some clinicians would use them for critically ill patients with lower respiratory tract disease. Convalescent plasma treatment has been used in patients with severe pandemic H1N1 and reduced mortality to 20% versus 55% in the non treatment group [57]. Human monoclonal antibodies targeting the extracellular domain of matrix protein 2 of influenza A virus are in development and might represent a more realistic therapeutic option than convalescent plasma [58].

Intravenous agents
Intravenous antiviral agents are potentially important because of issues involved with absorption of oral agents in the more severely ill patients. Oral agents might not be absorbed appropriately in patients with ileus or those with severe GVHD; aerosolized agents might not be delivered appropriately to the lungs in patients with acute respiratory distress syndrome or severe pneumonia. Two drugs have been used for intravenous therapy. Peramivir, a novel neuraminidase inhibitor, was available during the 2009 H1N1 pandemic under an emergency use authorization from the FDA for refractory cases and patients who could not tolerate oral medications [59]. It has been used in Japan and South Korea but remains investigational in most countries. Peramivir was effective in mouse studies against influenza A H1N1, H3N2, H5N1 as well as influenza B viruses [60,61]. Phase I and II trials had previously shown that peramivir was well tolerated in healthy adults. One study in 300 patients with acute uncomplicated influenza showed that peramivir significantly reduced time to alleviation of symptoms (P=0.0092) and a comparative study demonstrated that peramivir was non-inferior to oseltamivir (P<0.04) [62,63]. During the Emergency Investigational New Drug period, peramivir was administered to 20 adults and 11 children. Among those treated for 2–15 days, no serious adverse events were reported [64]. Thirty patients required mechanical ventilation; survival at 14 and 36 days after initiation of peramivir was 76.7% and 59.0%, respectively. Most patients had already failed a previous neuraminidase inhibitor and were critically ill before initiating peramivir. Only one HSCT recipient was included in this analysis. Clinical isolates of influenza A expressing the oseltamivir-resistant mutation H275Y in the neuraminidase have shown reduced susceptibility to peramivir [65,66]. Clinical failure of peramivir in HSCT recipients infected with H275Y mutant H1N1 has been described [67,68]. Peramivir is undergoing Phase III trials in hospitalized patients with influenza.

Intravenous zanamivir was shown to be safe and effective in 16 healthy volunteers inoculated with seasonal H1N1 and is undergoing Phase II evaluation [69]. This drug was used during the 2009 pandemic, generally in patients infected with oseltamivir-resistant H1N1 [68,70–79]. Of 10 cases in the literature, no severe adverse effects were reported although some patients died of respiratory failure. Viral load decrease after initiation of inhaled or intravenous zanamivir was shown in some cases but patients were critically ill and not all improved clinically. Successful treatment of oseltamivir-resistant cases with aerosolized or intravenous zanamivir has been reported (Figure 2) [79,80]. Nebulization of zanamivir aqueous saline solution, currently in development for intravenous administration, was well tolerated in 20 hospitalized patients with serious influenza [81]. However, even more limited data are available for patients undergoing mechanical ventilation and no data on effectiveness have been published. One case report of fatal respiratory events caused by ventilator filter blockade was reported when reconstituted lactose powder, meant for oral inhalation, was used for nebulization [82,83].

Antiviral resistance
Adamantane resistance conferred by S31N mutation in the M2 gene has been recognized to emerge rapidly in immunocompromised and eventually in immunocompetent patients infected with susceptible strains [84]. More recently, the S31N mutation was uniformly present in H3N2 strains and pandemic H1N1. Development of oseltamivir resistance in immunocompromised patients was described only rarely before 2007 [85,86]. Of five hematology-oncology patients infected with oseltamivir-resistant seasonal H1N1 circulating between 2007 and 2009, four had LRTI and three died of respiratory insufficiency [87,88]. Pandemic H1N1
Drug binds to respiratory epithelial cells and removes cell-surface sialic acid residues required by influenza virus to infect the cell [99,100]. DAS181 has shown in vitro inhibition of many strains of influenza viruses including oseltamivir- and zanamivir-resistant viruses [101–103]. In vitro selection of drug resistant mutant was minimal and fitness of the resistant viruses was considerably reduced [104]. Mouse studies have shown efficacy against different influenza strains and Phase II clinical trials are underway for treatment of community-acquired influenza with inhaled DAS181 [102,105]. DAS181 is available for compassionate use. Favipiravir (T-705) is a pyrazine derivative targeting the viral specific RNA-dependent RNA polymerase. This drug inhibits viral replication of influenza A, B and C viruses, including H5N1, drug resistant influenza, and pandemic 2009 H1N1 [106–110]. Combination therapy including oseltamivir and favipiravir has shown synergy and potentially better outcome in mouse studies [111]. Phase II clinical trials evaluating the efficacy and safety of oral favipiravir in adult patients with uncomplicated influenza is ongoing and enrolment in a Phase III study was recently completed in Japan. Lanimavir, a long acting neuraminidase inhibitor administered via a dry powder inhaler, was as effective as oseltamivir in a large double-blind randomized study [112]. The drug is potentially effective against oseltamivir-resistant virus and is currently available in Japan [113,114].

RSV

Initial studies performed on HSCT recipients with RSV pneumonia or LRTI reported mortality rates reaching 50–78% even with aerosolized ribavirin therapy [115–117]. Substantial progress in the diagnosis, supportive care, and targeting of the population at highest risk for severe RSV disease has been made over the past 20 years. RSV treatment modalities include ribavirin and RSV-specific monoclonal or polyclonal immunoglobulins. Ribavirin is a guanosine analogue available in aerosol, intravenous and oral formulations. Only one randomized controlled study was performed in HSCT recipients, showing that preemptive aerosolized ribavirin was safe and could potentially decrease viral load over time [118]. Although LRTI developed in none of 9 patients treated with ribavirin and 2 of 5 untreated patients, no conclusion on clinical efficacy could be made. One recent retrospective analysis among adult HSCT population based on uncontrolled studies showed that preemptive treatment of URTI with 5–7 days of inhaled ribavirin decreased progression to LRTI from 47% to 25% (P=0.01) [119]. Addition of RSV-specific monoclonal or polyclonal immunoglobulins reduced the progression rate from 45% to 12% (P<0.001). Treatment of established LRTI is more challenging because of the associated high mortality. The
same investigators compared mortality rates of 89% in untreated patients compared to 50% in patients treated with inhaled ribavirin (P<0.04). The addition of RSV-specific monoclonal or polyclonal immunoglobulins to inhaled ribavirin reduced mortality from 77% in untreated patients to 24% (P<0.001). Fewer data are available in children but 10 of 11 paediatric HSCT recipients with RSV LRTI survived when treated with ribavirin and RSV-IVIG [120].

Immunocompromised patients at our institution with RSV URTI or asymptomatic shedding are treated with 10 days of inhaled ribavirin if they have severe lymphopenia (absolute lymphocyte count ≤300/mm³). Patients with lymphocytes >300/mm³ are usually only observed closely. For RSV LRTI, patients receive 10 days of inhaled ribavirin in addition to 15 mg/kg of palivizumab or 400 mg/kg of IVIG, since RSV-IVIG is not routinely available. Even if the addition of RSV-specific immunoglobulins to ribavirin were more effective than ribavirin alone in reducing the progression from URTI to LRTI according to recent retrospective analysis, the additional cost of the RSV-specific immunoglobulins requires more supportive data.

Limited data on treatment with intravenous and oral ribavirin for RSV is available. Intravenous ribavirin would be appealing in patients with severe lung disease where distribution of inhaled ribavirin might be suboptimal, or ventilator support is required. Intravenous ribavirin is usually well tolerated but possible side effects include haemolysis, leukopenia and hyperbilirubinemia [121]. Mortality in studies with small numbers of patients with RSV LRTI treated with intravenous ribavirin ranged from 33% to 80% [17,121,122]. Oral ribavirin could potentially be useful to treat patients who do not need hospitalization, reducing the complexity of therapy with inhaled ribavirin and the theoretical risk of teratogenicity in care givers. Although oral ribavirin appears to be safe, reversible anaemia was a common side effect [123]. Uncontrolled studies of serial nasal secretions from 19 patients treated with oral ribavirin revealed a decrease in RSV load of 2 log₁₀ copies/ml within 7 days after initiation of therapy in 58% of the patients and within 14 days after treatment initiation in 90% of the patients [124]. Progression to LRTI and mortality associated with LRTI were similar to studies using aerosolized ribavirin (14% and 43%, respectively). Another study using oral and/or intravenous ribavirin did not report any progression to LRTI among treated patients with URTI; mortality among treated patients with LRTI was similar to published mortality in patients treated with inhaled ribavirin (5/12 patients; 42%) [125]. Six paediatric hematology-oncology/HSCT patients have received oral ribavirin plus IVIG for URTI or LRTI without adverse events or death [126].

Investigational agents

The second generation RSV-specific monoclonal immunoglobulin, motavizumab, has been investigated for more than 5 years. This new monoclonal immunoglobulin also targets the antigenic site A on the fusion (F) glycoprotein of RSV but with a neutralizing activity 18-fold greater than palivizumab in vitro and up to 100-fold in the cotton rat-model [127,128]. Phase III studies in preterm infants reported motavizumab as non-inferior to palivizumab in RSV prophylaxis [129,130]. However, the FDA did not approve this drug in 2010, and the manufacturer discontinued most motavizumab development and withdrew the paediatric prophylaxis application. A Phase II study is ongoing for RSV treatment in children ≤12 months hospitalized with RSV LRTI. No data is available in HSCT recipients.

To facilitate the development of new drugs targeting RSV, the relationship between viral load and disease in healthy adults experimentally infected with RSV has been studied [131]. This study demonstrated that RSV disease is related to the virus replication measures in healthy young adults, and thereby supports a potential benefit of RSV antivirals. Subsequently, two randomized clinical trials reported the safety and potential efficacy of aerosolized ALN-RSV01, a small interfering RNA (siRNA) targeting RSV replication. The first study showed reduction of RSV infection by 44% compared to placebo in experimentally infected adults receiving ALN-RSV01 prophylaxis [132]. The second study, conducted in lung transplant recipients, showed lower symptom scores and a decrease of new or progressive bronchiolitis obliterans (6.3% versus 50%; P=0.027) with ALN-RSV01 treatment [133]. A larger Phase IIb study is ongoing in lung transplant recipients. No data are available in HSCT recipients and the drug is not yet available for compassionate use.

Human adenovirus

In the last decade, HAdV infections have been increasingly recognized as a major cause of morbidity and mortality among HSCT recipients. The pathogenesis of adenovirus disease is not well understood but the presence of CD4 and CD8 lymphocytes has been essential for viral clearance. The incidence of HAdV is highly variable, with highest rates in paediatric HSCT recipients compared to adults (24–32% versus 3–9%) [134–136]. Disseminated disease is less frequent and has been reported in 1–7% of HSCT recipients with an associated mortality of 80%. Adenovirus-DNAemia that is rapidly increasing or steadily high is associated with severe disease in children and adults [137–139]. Literature suggesting weekly monitoring of HAdV-DNAemia in high-risk paediatric population is increasing [134,135,138,140]. However, the usefulness of
monitoring in adult allo-HSCT recipients has not been shown [136,141]. HAdV monitoring may be beneficial in the highest risk adult population including refractory GVHD, umbilical cord blood transplantation, haploidentical transplantation, stem cell graft depletion over 2–3 log10 T-cells and use of anti-T-cell antibodies. HAdV disease onset is usually around 30 days post-transplantation in children and tends to be closer to 100 days in adult HSCT recipients. Detection of HAdV in stool by real-time PCR precedes HAdV-DNAemia and could provide earlier start of preemptive treatment and improve outcome [142]. The presence of HAdV in multiple sites (blood, stool, urine, respiratory samples) correlates with progression to disseminated disease [138,143]. HAdV positivity in nasopharyngeal aspirate prior to HSCT in paediatric patients is also strongly predictive of HAdV-DNAemia, potentially guiding the timing of elective HSCTs or targeting specific patients for preemptive treatment [144].

Cidofovir, a monophosphate nucleotide analogue of cytosine, is the agent with the most evidence for efficacy against HAdV. Cidofovir is effective against all strains of HAdV in vitro and resistance has only rarely been described after serial passage in cell culture [145]. Mutations in the DNA polymerase gene were possibly associated with cidofovir resistance, similar to that seen in cytomegalovirus, herpes simplex virus and human herpesvirus type 6 [146–148], but no clinical case of cidofovir-resistant HAdV has yet been described [149]. Cidofovir can be used as preemptive or therapeutic treatment. Given the high mortality rate associated with adenovirus disease and the low efficacy of antivirals in disseminated disease, preemptive treatment is often the favoured option [150].

No randomized clinical trials evaluating preemptive treatment strategies have been performed, with available data coming only from retrospective and prospective cohort studies. These studies are limited by the lack of control and the incompletely understood natural history of asymptomatic adenovirus disease in HSCT patients. One of the largest studies treated 14 asymptomatic patients and 43 symptomatic patients, mostly children. None of the 14 asymptomatic patients developed disease and only one of the 43 symptomatic patients died of adenovirus disease [151]. Another prospective study screened 38 paediatric HSCT recipients and identified seven with high-risk HAdV infections. All seven patients received preemptive cidofovir therapy and cleared their infection without complications [134]. A retrospective study in 23 patients, mostly children, reported that cidofovir therapy was well tolerated in 87% of the patients and was considered successful in 85% of patients [152]. One meta-analysis identified 141/181 patients (77%) who were treated successfully with cidofovir monotherapy based on disease resolution, survival and clearance of the virus for 7 days after discontinuation of cidofovir [152]. However, that group included both symptomatic and asymptomatic patients. Another retrospective study analysed the outcome of 20 paediatric HSCT patients infected with HAdV pre- and post-implementation of a clinical algorithm. The study reported that 4 out of 5 high-risk untreated patients (prealgorithm) died secondary to HAdV infection compared to 1 of 9 high-risk treated patients (postalgorithm), decreasing the mortality rate from 80% to 11% (P<0.05). Importantly, monitoring was not performed consistently in the prealgorithm phase, thereby potentially selecting for sicker patients. A detailed algorithm for HAdV treatment in HSCT recipients has been published recently [153].

Different cidofovir dosages have been assessed to reduce the nephrotoxicity associated with therapy (Table 2). Regimens including 1 mg/kg, three times a week is considered less nephrotoxic than the usual dose of 5 mg/kg once a week. This lower dose was shown to be effective in prevention of disease [134]. Hyperhydration and coadministration of probenecid, 3 h before cidofovir administration, decrease nephrotoxicity. Cidofovir causes renal proximal tubular cells dysfunction and leads to proteinuria in 50% of reported case, increased serum creatinine levels in 9–25% of HSCT recipients and Fanconi syndrome in 1% of patients [152,154,155]. Rare cases of chronic interstitial nephritis and nephrogenic diabetes insipidus have also been described [135]. Duration of therapy is based on drug tolerance and clearance of the virus which can take many weeks.

Alternative therapeutic options for the treatment of adenovirus disease include intravenous ribavirin which is effective in vitro against subgroup C and possibly other subgroups depending on the 50% inhibitory concentration [149,156]. Clinical data using ribavirin have been conflicting, showing no or minimal efficacy. One study looking at viral loads after initiation of ribavirin in four HSCT recipients did not show any response even when the infection was caused by subgroup C viruses [157]. There is scant data supporting ribavirin as primary therapy for HAdV infections but this might be considered in specific situations or as adjunct therapy. Ganciclovir is also effective in vitro against HAdV but limited data exist on efficacy of ganciclovir in the treatment of clinical disease. Two studies have revealed that patients receiving ganciclovir prophylaxis were at lower risk of HAdV infection and disease than patients who did not [158,159]. Whether ganciclovir has an effect on HAdV is unclear; more data is required. IVIG has been utilized with success in a small number of patients but no recommendations can be made without additional data [160–162].
### Table 2. Antiviral dose and toxicity

<table>
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<th>Drugs</th>
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<th>Doses</th>
<th>Adverse events</th>
<th>Suggested monitoring</th>
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</thead>
<tbody>
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<td>Oseltamivir</td>
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<td>Treatment if $&gt;40$ kg: 150 mg PO twice a day Treatment if $&lt;40$ kg: 75 mg PO once or twice a day $\times$10 days</td>
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<td>Zanamivir</td>
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<td>Adult and paediatric: 10 mg inh twice a day $\times$5 days Same dose for prophylaxis and treatment</td>
<td>Bronchospasm GI Neuropsychiatric</td>
<td>NA</td>
</tr>
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<td>Peramivir</td>
<td>Dose adjustment for renal function</td>
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<td>GI Neutropenia</td>
<td>CBC Biochemistry Liver function Renal function Urinalysis</td>
</tr>
<tr>
<td>Amantadine</td>
<td>Dose adjustment for renal function</td>
<td>$&lt;10$ years old: 5 mg/kg/day PO 1–2 divided doses, max 150 mg/day $&gt;10$ years old 100 mg PO twice a day</td>
<td>Nervousness Anxiety Lightheadedness Seizure Ataxia</td>
<td>Renal function prior initiation Close neurological follow-up in older patients</td>
</tr>
<tr>
<td>Rimantadine</td>
<td>Caution in patient $&gt;65$ years old or with CNS disease or psychosis or with concomitant anticholinergic drug therapy</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inhaled ribavirin</td>
<td>Risk of teratogenicity for 6 months after therapy Consider concomitant administration of a bronchodilator</td>
<td>Adult and paediatric: 2 g/dose 3 times a week or 6 g over 12–18 h</td>
<td>Bronchospasm</td>
<td>Attention must be directed to procedures that minimize accumulation of drug precipitate, which can result in mechanical ventilator dysfunction and associated increased pulmonary pressure</td>
</tr>
<tr>
<td>Oral ribavirin</td>
<td>Dose adjustment for renal function Risk of teratogenicity for 6 months after therapy</td>
<td>If $&gt;40$ kg: 10 mg/kg PO as loading dose, 400 mg three times a day on day 2 and 600 mg three times a day thereafter for adenovirus 10–20 mg/kg PO four times a day If $&lt;40$ kg: 2 mg/kg PO three times a day</td>
<td>Anaemia</td>
<td>CBC</td>
</tr>
<tr>
<td>IV ribavirin</td>
<td>Dose adjustment for renal function Risk of teratogenicity for 6 months after therapy</td>
<td>General dosage: 33 mg/kg loading dose, followed 6 h later by 16 mg/kg every 6 h for 4 days (total of 36 doses), then 8 mg/kg every 8 h for 3 days (total of 9 doses)</td>
<td>Haemolytic anaemia Leukopenia Hypocalcaemia Elevated ALT</td>
<td>CBC Biochemistry Liver function</td>
</tr>
</tbody>
</table>

ALT, alanine aminotransferase; CBC, complete blood count; CNS, central nervous system; GI, gastrointestinal; IgA, immunoglobulin A; inh, inhaled; IV, intravenous; IVIG, pooled intravenous immunoglobulins; NA, not applicable; PO, per os.
<table>
<thead>
<tr>
<th>Drugs</th>
<th>Contraindication/precautions</th>
<th>Doses</th>
<th>Adverse events</th>
<th>Suggested monitoring</th>
</tr>
</thead>
<tbody>
<tr>
<td>Palivizumab</td>
<td>NA</td>
<td>15 mg/kg/dose IV</td>
<td>NA</td>
<td>IVIG Do not administer if patient 0.5 g/kg</td>
</tr>
<tr>
<td>Cidofovir</td>
<td>Dose adjustment for Adult and paediatric: 3–5 mg/kg IV</td>
<td>Cidofovir: Nephrotoxicity Monitor within 48 h prior to renal function one time a week or 1 mg/kg IV 3 times a week (tubulopathy, acute renal failure, Fanconi syndrome, Renal function is known for IgA deficiency)</td>
<td>Anaphylaxis</td>
<td>CBC during and after therapy Contraindication is recommended other nephrotoxic agents Not recommended with renin function Cidofovir is known for IgA deficiency Close follow-up for anaphylaxis</td>
</tr>
</tbody>
</table>

Treatment of adenovirus infections: severe infections in immunocompromised children

| HAART | 1 g 2 h after and 1 g 8 h after | 7 g/day or 0.5 g/kg PO 3 times a day 72 h before cidofovir infusion, followed by 1 g 2 h after and 1 g 8 h after | Neutropenia because of teratogenicity Hemolytic anaemia (G6PD deficiency) | 15 mg/kg/dose IV 15 mg/kg/dose IV 75 mg/kg/day IV twice daily in children infections in immunocompromised children |

Table 2. Continued
Investigational agents
CMX001 is an orally available lipid conjugate of cidofovir not affected by compromised renal function and not associated with nephrotoxicity [163]. Phase I studies have not shown any major toxicity and studies in immunosuppressed Syrian hamsters have shown efficacy against HAdV infection [164]. A retrospective analysis of 13 immunocompromised patients treated with CMX001 for HAdV-DNAemia reported significant decrease in viral loads, a complete response rate of 69.3% and no serious adverse events [165]. A pediatric HSCT recipient with disseminated HAdV infection was successfully treated with CMX001 and his renal function improved [166]. An open-label study for the treatment of dsDNA virus infection, including HAdV, is underway as well as a Phase II study for specific treatment of HAdV infection in paediatric and adult HSCT recipients.

Effective T-cell function seems to be crucial for control of adenovirus replication [167–170]. Therefore, reduction of immunosuppression may improve T-cell function and outcome. Exogenous lymphocyte therapy is promising and several protocols have been published [170–173]. One trial in paediatric HSCT recipients using ex vivo isolated interferon-g-producing T-cells reported decreased viral loads in 5 of 6 patients [171]. Implementation of immunotherapy for HAdV infections is confined to highly specialized centres. Improvement in timeliness and cost-effectiveness will be necessary to offer immunotherapy for clinical care.

Parainfluenza
Parainfluenza infections in HSCT recipients result in pneumonia with severe disease more often when patients receive systemic corticosteroids or have lymphopenia [20]. No drug has shown benefits in controlled studies, although in vitro and in vivo data have shown efficacy of ribavirin against PIVs [174]. Inhaled, oral or intravenous ribavirin formulations have been used to treat parainfluenza infection in HSCT recipients. One retrospective analysis reported that neither inhaled ribavirin nor IVIG improved the outcome of HSCT recipients with PIV3 pneumonia [20]. Other case reports and series have commented on potential efficacy of ribavirin against PIV [175–178]. Two studies looked at preemptive treatment of PIV URTI but because of small numbers and absence of controls, no recommendation can be made [179,180]. Some benefits may have been seen in PIV-infected lung transplant recipients treated with ribavirin [181,182].

DAS181, a recombinant sialidase protein discussed above, is also active against PIV. The demonstration of anti-PIV activity for DAS181 in a culture model reflecting human airways and in animal models has triggered substantial interest [183]. Inhaled DAS181 was administered for three days to a PIV3-infected HSCT recipient with a reduction of nasopharyngeal viral load observed. By day three, the patient no longer required supplemental oxygen. In vitro testing confirmed the strain susceptibility to DAS181 [184]. The drug is available for compassionate use in severe PIV infections. Another new drug, BCX 2798, is a haemagglutinin-neuraminidase inhibitor with high efficacy against PIV1 in vitro and in vivo as prophylaxis and therapeutically [185,186]. Studies in humans are not available.

Human metapneumovirus
Ribavirin is active in vitro and in vivo against HMPV but no drug has shown clinical effectiveness [187,188]. Intravenous ribavirin and IVIG have been reported as potentially successful therapeutic options [189–191]. A retrospective analysis compared the outcome between 13 immunocompromised patients with HMPV pneumonia treated with ribavirin ±IVIG and 10 untreated patients. Ribavirin treatment was associated with more hypoxaemia and similar mortality, possibly related to late initiation of therapy [192].

Other viruses
The importance of rhinovirus and coronavirus requires further study in HSCT recipients. Severe respiratory disease with both viruses has been described with no specific agent available for treatment [14,15]. Chemotherapeutic approaches targeting picornavirus have been the object of intensive research. Pleconaril, a capsid-binding agent, was the most promising of all agents. Pleconaril decreased by 1 day the median time to alleviation of illness among 1,363 picornavirus-positive subjects compared to placebo [193]. Subsequently, baseline and acquired resistance was described [194]. No experience with pleconaril in HSCT patients has been described and the drug is not available for compassionate use. More recently, another capsid-binding agent named BTA-798 has completed Phase I and Phase IIa studies, achieving clinical proof-of-concept by reducing the incidence and severity of infection as well as viral load in healthy volunteers. Another Phase II study is actually recruiting patients with asthma to evaluate efficacy and safety of BTA-798. Inhaled interferon-β 1a (SNG001) is another compound in development to treat rhinovirus in asthmatic patients by improving anti-viral response of the respiratory epithelium. Phase II study of this drug is also ongoing in asthmatic patients. The effect of these potential drugs in HSCT recipients is difficult to predict. The epidemiology and clinical effect of other emerging viruses (HBoV, polyomavirus WU and KI) are not yet known and no treatment has been suggested.
Conclusion

Progress in understanding the epidemiology and treatment of respiratory viruses has greatly increased over the last decade. However, treatment recommendations for respiratory viral infections in HSCT recipients are based mainly on retrospective and prospective cohort studies. Determination and standardization of viral loads in clinical samples with new molecular assays may be useful to document antiviral activity, assess the importance of viral loads in disease, and assist in the development of new drugs. Larger studies evaluating treatment of PIV and HMPV infections are urgently needed as well as studies looking at the effect of treatment on long-term complication.

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References


Antiviral therapy of respiratory viruses in HSCT recipients


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