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New Developments in the Management of Cytomegalovirus Infection after Solid Organ Transplantation

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Abstract

Despite remarkable advances in the diagnostic and therapeutic modalities for its management, cytomegalovirus (CMV) remains one of the most important pathogens impacting on the outcome of transplantation. Not only does CMV directly cause morbidity and occasional mortality, it also influences many short-term and long-term indirect effects that collectively contribute to reduced allograft and patient survival. Prevention of CMV infection and disease is therefore key in ensuring the successful outcome of solid organ transplantation (SOT).

In this regard, antiviral prophylaxis and pre-emptive therapy are similarly effective in preventing CMV disease after transplantation. However, current guidelines prefer antiviral prophylaxis over pre-emptive therapy in preventing CMV disease in high-risk SOT recipients, such as CMV-seronegative recipients of organs from CMV-seropositive donors (CMV D+/R−), and lung, intestinal and pancreas transplant recipients. Antiviral prophylaxis has the benefits of reducing not only the incidence of CMV disease, but also the indirect effects of CMV on allograft and patient survival. The major

drawback of antiviral prophylaxis is delayed-onset CMV disease, which occurs in 15–38% of CMV D+/R– SOT recipients who received 3 months of prophylaxis. Allograft rejection, over-immunosuppression and lack of CMV-specific immunity are factors that predispose patients to delayed-onset CMV disease. A recent randomized trial in CMV D+/R– kidney recipients demonstrates a significant reduction in the incidence of CMV disease when valganciclovir prophylaxis is extended to 200 days (compared with the standard 100 days) after transplantation; however, the safety and cost of this prolonged approach has yet to be assessed. In some studies, delayed-onset CMV disease has been significantly associated with allograft loss and mortality. In the vast majority of patients, CMV disease responds to treatment with intravenous ganciclovir. Recently, oral valganciclovir was demonstrated to have an efficacy that is comparable to intravenous ganciclovir in treating mild to moderate cases of CMV disease in SOT recipients. Reduction in the degree of immunosuppression should complement antiviral treatment of CMV disease.

Although it remains rare, ganciclovir-resistant CMV disease is increasingly seen in clinical practice, potentially fostered by the prolonged use of antivirals in high-risk over-immunosuppressed transplant recipients. Treatment of drug-resistant CMV is currently non-standardized and may include foscarnet, cidofovir, CMV hyperimmune globulins or leflunomide. The investigational drug marivabir had the potential to treat ganciclovir-resistant CMV disease as it acts through a different mechanism. However, the recent phase III clinical trial in allogeneic bone marrow transplant recipients showed that maribavir was not significantly better than placebo for the prevention of CMV disease. Similarly, the preliminary data in a liver transplant population suggests that maribavir was inferior to oral ganciclovir for the prevention of CMV disease. This article reviews the recent data and other developments in the management of CMV infection after SOT.

Cytomegalovirus (CMV) continues to be the single major pathogen affecting the outcome of solid organ transplantation (SOT).[1] Transplant recipients develop CMV disease either as a primary infection (when CMV is transmitted through the transplanted allograft to a CMV-naive SOT recipient), a reactivation infection (when an endogenous latent virus in the SOT recipient reactivates after transplantation) or as reinfection (when donor-transmitted virus is superimposed on an endogenously reactivated virus). Of these three mechanisms, primary CMV infection is considered to be the most serious in terms of direct clinical disease and outcome. Not only does CMV directly cause morbidity and occasional mortality, it also influences many short-term and long-term indirect effects that collectively contribute to reduced allograft and patient survival (table I).[2,3]

The measures that are currently used for the prevention of CMV infection and disease in SOT recipients – antiviral prophylaxis and pre-emptive therapy – have reduced the direct effects of this infection. However, the morbidity associated with CMV remains, particularly among high-risk transplant populations.[4] Despite antiviral prophylaxis among high-risk SOT recipients, particularly CMV-seronegative recipients of organs from CMV-seropositive donors (CMV D+/R–), a large number (estimates range from 15% to 38%) still develop delayed-onset (or late-onset) CMV disease soon after the completion of antiviral prophylaxis.[3,5,6] Although others have suggested that late-onset CMV disease is not a major factor that influences transplant outcomes,[7] there is an increasing amount of data to implicate delayed-onset CMV disease as a harbinger for higher rates of allograft failure and mortality.[2,3] These latter

Table I. Clinical impact of cytomegalovirus (CMV) after solid organ transplantation

Direct effects
CMV syndrome
fever, myalgia, arthralgia
myelosuppression
Tissue-invasive CMV disease
gastrointestinal disease
hepatitis
pneumonitis
CNS disease
retinitis
nephritis
pancreatitis
carditis
others[a]
Mortality
Indirect effects
Acute rejection
Chronic rejection
bronchiolitis obliterans
transplant vasculopathy
tubulointerstitial fibrosis
Immunosuppression with opportunistic infections
fungal infections
bacterial infections (nocardiosis, listeriosis)
viral infections (HHV-6, HHV-7, HCV)
viral-associated neoplasia (PTLD)
New-onset diabetes mellitus
Mortality

a Any organ system may be affected by CMV.

HCV = hepatitis C virus; **HHV** = human herpesvirus; **PTLD** = post-transplant lymphoproliferative disease.

observations should highlight the need for better preventive strategies and this continues to be the major focus of ongoing clinical investigations. In this article, we review the current state of CMV management in SOT recipients, with particular emphasis on the recent developments in the field.

1. Traditional and Newly Identified Risk Factors of Cytomegalovirus (CMV) Disease after Transplantation

There are numerous factors that contribute to the common occurrence of CMV infection after SOT. In general, CMV infection occurring in an environment of markedly impaired or deficient immunity is the scenario that translates to the occurrence of CMV disease. The classic situation is the CMV D+/R− sero-mismatched SOT recipient, wherein CMV reactivation (in the transplanted allograft) and replication occurs in a patient who does not possess pre-existing CMV-specific immunity. Lack of pre-existing immunity will allow for CMV replication to remain unabated, especially in pharmacologically immunosuppressed SOT recipients, thereby leading to symptomatic clinical disease. Accordingly, a CMV D+/R− serostatus is considered to be the most important risk factor for the development of CMV disease after SOT. Without an active prevention strategy, most CMV D+/R− SOT patients will develop primary CMV infection and disease after transplantation. Antiviral prophylaxis has reduced the incidence of CMV disease in CMV D+/R− SOT patients. However, despite the protection afforded during the time of primary prophylaxis, most of the patients fail to develop sufficient CMV-specific immunity to allow efficient control of viral replication upon the completion of prophylaxis, which predisposes them to develop delayed-onset (or late-onset) primary CMV disease.[4] Recent data suggests that individuals deficient of CMV-specific T-cell immunity were more likely to develop delayed-onset CMV disease (table II).[8]

Deficiency in T-cell mediated immunity is heightened by the use of induction immunosuppressive therapy, especially of lymphocyte-depleting agents such as muromonab-CD3 (OKT3) and antithymocyte globulin[4] or alemtuzumab,[9,10] thereby increasing the subsequent risk of CMV disease. These immunosuppressive agents cause profound global depletion of T lymphocytes for a considerable period of time, thereby failing to control the replication of a reactivated virus. A similar scenario exists when SOT patients receive a more intense degree of maintenance immunosuppression, as exemplified by the clinical observation of a higher risk of CMV disease with the use of a three-drug rather than a two-drug immunosuppressive regimen.[11] The use of mycophenolate mofetil and prednisone at the end of antiviral prophylaxis was

Table II. Traditional and newly-identified risk factors for cytomegalovirus (CMV) disease after solid organ transplantation

CMV serostatus of the donor (D) and recipient (R)
CMV D+/R– > D+/R+ > D–/R+ > D–/R–
Allograft rejection
Active CMV infection
high viral replication
Type of organ transplant
lung > heart > kidney
Maintenance immunosuppressive therapy
mycophenolate mofetil
Induction immunosuppressive therapy
muromonab-CD3, antithymocyte globulin, alemtuzumab
Concomitant viral infections
HHV-6, HHV-7
T-cell anergy
lack of CMV-specific CD4+ and CD8+ T cells
Genetic polymorphism of the innate and adaptive immune system
TLR 2, TLR 4, CCR 5, mannose binding lectin, IL-10, MCP-1
Renal insufficiency
Others
retransplantation, volume of blood transfusion, sepsis and other factors associated with high TNFα secretion

CCR 5 = chemokine (C-C motif) receptor 5; **HHV** = human herpesvirus; **IL-10** = interleukin-10; **MCP-1** = monocyte chemotactic protein-1; **TLR** = Toll-like receptor; **TNFα** = tumour necrosis factor-α.

associated with increased risk of CMV disease in liver transplant recipients (relative to the group of patients who have been successfully tapered off of these drugs and maintained on tacrolimus monotherapy).[5,12]

One major reason for an over-immunosuppressed state is the need to treat or aggressively prevent acute rejection, which by itself has been associated with CMV disease in SOT recipients.[13] There is a bidirectional relationship between CMV disease and acute rejection after transplantation. Through immunomodulation and upregulation of alloantigens, CMV promotes the development of acute rejection. Conversely, allostimulation and acute rejection can transactivate CMV, which is further aggravated by use of anti-rejection treatment, especially with lymphocyte-depleting agents.[9] These observations have led to the standard practice of using anti-CMV prophylaxis concomitantly with treatment of acute rejection.[3]

As efficient CMV-specific immunity is a major player in achieving successful control of CMV, T-cell assays may potentially be useful in assessing the risk of CMV disease in transplant recipients.[14-17] However, the results of studies evaluating the association between CMV-specific T cells and clinical outcomes have so far been conflicting. In a study of 38 SOT recipients, early immunological responders with T-cell restoration during the first month following transplantation were able to control CMV even without antiviral treatment, while late responders often required antiviral therapy.[17] In another study, patients with detectable CMV-reactive CD4+ T cells during the first month after cardiac transplantation were less likely to have CMV replication, acute rejection and vasculopathy.[16] In a third study, the detection of interferon (IFN)-γ during stimulation of CD8+ T cells with CMV peptides was correlated with a significantly lower risk of late-onset CMV disease.[8] On the other hand, the presence and the level of CMV-specific CD4+ and CD8+ T cells were not significantly associated with subsequent CMV DNAaemia in a study of 45 kidney transplant recipients who received valganciclovir prophylaxis.[18] This concurs with previous studies showing no significant correlation between CMV-specific CD4+ and CD8+ T cells and the development of subsequent CMV infection. In a study of 20 kidney transplant recipients, a high CD8+ T-cell response was found to be the 'consequence', rather than 'predictor', of CMV viraemia.[14] Similarly, another study found no correlation between CMV viraemia and the levels of IFNγ-producing CD8+ T cells in CMV D+/R– liver recipients.[19] The disparities in the results of these studies could be due to the differences in patient populations being studied (CMV-seropositive vs CMV D+/R– patients and type of immunosuppressive regimens) and in the assays that are used (cytokine flow cytometry, IFNγ release, enzyme-linked immunosorbant spot [ELISPOT] assays). Currently, these assays of CMV-specific immunity are non-standardized and require further development and optimization, and hence cannot yet be used routinely in clinical practice to guide preventive and therapeutic strategies.

Components of the innate immune system are also involved in the control of CMV infection after transplantation, as suggested by studies investigating various aspects of the innate immune system. Single nucleotide polymorphisms in the Toll-like receptor (TLR)-2 gene have been associated with increased CMV replication and disease after liver transplantation,[20] while TLR4 gene polymorphism was associated, albeit marginally, with CMV disease after kidney transplantation.[21,22] Mannose binding lectin (MBL) deficiency has also been associated with CMV infection or disease in a small study of kidney transplant recipients.[22,23] In this study, MBL deficiency was present in five of seven (71%) patients with CMV disease, all four (100%) patients with CMV infection, but not in five patients without CMV infection ($p = 0.005$).[23] Finally, polymorphisms of genes encoding for the chemokine (C-C motif) receptor 5, interleukin-10 and monocyte chemotactic protein-1 have also been associated with CMV disease after transplantation.[24]

2. Clinical Impact of CMV in Transplant Recipients

CMV infection in SOT recipients can either be asymptomatic or progress to full-blown clinical disease. One of the major factors that determine this clinical presentation is the net state of immunosuppression in the host. Transplant patients who are receiving a higher degree of immunosuppression are more likely to develop clinical disease, as they are less likely to mount an effective immune protection against the virus.

2.1 Direct Effects

Most symptomatic patients will manifest with a CMV syndrome characterized by nonspecific signs and symptoms such as fever, fatigue, body aches and myelosuppression. In some patients, CMV disease is manifested as tissue-invasive disease. The gastrointestinal tract is the most common site of predilection for tissue-invasive CMV disease, independent of the type of allograft transplant.[25] In this regard, patients often present with abdominal pain and diarrhoea. In severe cases, CMV ulceration in the gastrointestinal tract could lead to haemorrhage and perforation. Other organs that could be involved include the liver, lungs, heart, pancreas and kidneys, and may manifest with allograft dysfunction that could be misdiagnosed as acute or chronic rejection (table I).[4] For unclear reasons, chorioretinitis, which is a common manifestation of CMV disease in AIDS patients, is a very rare clinical presentation in SOT recipients.[26]

2.2 Indirect Effects

CMV is associated with a variety of indirect effects that are mediated by its ability to modulate the immune system (table I).[4] SOT recipients with CMV infection or disease are more likely to develop opportunistic infections due to other viruses (i.e. human herpesvirus [HHV]-6, HHV-7, Epstein-Barr virus-related post-transplant lymphoproliferative disease),[27-29] bacteria (e.g. *Nocardia* spp.)[30] and fungi.[31] CMV infection was associated with poor allograft and patient outcome due to hepatitis C virus recurrence in liver recipients,[32] although this was not demonstrated in another study.[33] Aside from infections, patients with CMV infection are more likely to experience acute and chronic rejection. Transplant vasculopathy in heart transplant recipients and bronchiolitis obliterans in lung transplant recipients continue to be reported, most notably in cases of suboptimal CMV preventive strategies.[34-37] CMV seropositivity of cardiac recipients and the lack of aggressive anti-CMV prophylaxis were significantly correlated with negative vascular remodelling and greater loss of vascular lumen.[34] In one study, pre-transplant recipient CMV seropositivity was a more powerful predictor of vasculopathy and death after heart transplantation than traditional risk factors.[35] CMV infection has also been described as an independent risk factor for atherosclerosis in kidney recipients.[36] Interestingly, new-onset diabetes mellitus has also been reported in patients with CMV infection or disease after kidney transplantation,[38,39] while the development of γ-δ T cells in response to CMV has been reported

to be associated with a lower risk of malignancy after kidney transplantation.[40]

CMV infection is associated with an overall higher rate of allograft failure and death of SOT recipients, in part, due to increased opportunistic infections and acute and chronic allograft rejection.[4,41] In one study, CMV persistence in the allograft was associated with reduced allograft function and survival after kidney transplantation.[42] Furthermore, late-onset CMV disease was found to be a strong and independent predictor of mortality in liver transplant recipients.[2] The indirect effects of CMV, particularly allograft rejection and vasculopathy, have been minimized by strategies of CMV prevention.[43-45] However, primary antiviral prophylaxis appears to be more effective in preventing the indirect effects of CMV than pre-emptive therapy.[46,47]

3. Prevention of CMV Disease in Recipients of Solid Organ Transplantation (SOT)

The two major antiviral strategies for the prevention of CMV disease after SOT are antiviral prophylaxis and pre-emptive therapy. Antiviral prophylaxis is the strategy wherein an antiviral drug is provided to all patients at risk of CMV disease beginning at the time of transplantation and lasting for at least 3 months. In contrast, pre-emptive therapy is the strategy wherein patients are monitored for evidence of CMV reactivation by use of CMV nucleic acid amplification assay or antigenaemia assay and, as soon as CMV is detected, antiviral drugs are given with the goal of preventing the progression of the infection into full-blown clinical disease.

3.1 Antiviral Prophylaxis

Antiviral prophylaxis is recommended for all high-risk SOT recipients, including CMV D+/R− SOT recipients, lung/heart-lung recipients, intestinal recipients and pancreas/kidney-pancreas recipients.[48] Using this strategy, all high-risk SOT patients receive antiviral medications for the first 3–6 months (or even longer) following transplantation.[48] Multiple clinical trials have demonstrated the benefits of antiviral prophylaxis in reducing the incidence of CMV disease. In recent meta-analyses that collectively compiled the results from individual clinical trials, it was demonstrated that primary antiviral prophylaxis was effective in preventing the direct and some of the indirect effects of CMV (table III).[46,47,49] Compared with placebo or no treatment, primary CMV prophylaxis reduced the incidence of CMV disease by 58–80%[46,47,49] and CMV infection by 39%.[46] Further investigations of the indirect effects of CMV showed that the incidence of acute rejection was reduced by 25%.[47] In addition, infections due to other herpesviruses, bacteria and protozoa were less common among patients who received antiviral prophylaxis.[46] Moreover, all-cause mortality was reduced[46,47] and this was mainly due to the decline in CMV-related deaths.[46]

3.1.1 Drugs Used for CMV Prophylaxis

Over the years, there have been a variety of antiviral agents used for CMV prophylaxis, with the continual development of newer agents that are intended to be better than previous drugs. Currently, the drugs with proven efficacy for the prevention of CMV disease are valaciclovir (in kidney transplant recipients only), oral and intravenous ganciclovir, and oral valganciclovir, a prodrug of ganciclovir with much improved bioavailability (table IV). Foscarnet and cidofovir are generally not regarded as first-line drugs for the prevention of CMV disease due to concerns about their toxicity profile, primarily nephrotoxicity in patients who are already receiving other drugs that could potentially affect renal function. An alkoxyalkyl prodrug of cidofovir, CMX-001 (also known as HDP-cidofovir, where HDP is hexadecyloxy-propyl), is anticipated to have a lower toxicity profile and is currently being evaluated in early-phase clinical trials as an oral agent for the prevention of CMV disease in transplant recipients.[51] Of all the drugs currently available in the clinical setting, valganciclovir is considered as the drug of choice in most transplant centres because of its excellent pharmacokinetic profile.

Antiviral prophylaxis with valganciclovir (900 mg once daily) was compared with oral

Table III. Recent meta-analyses of randomized controlled trials (RCTs) of antiviral prophylaxis and pre-emptive therapy for the prevention of cytomegalovirus (CMV) disease after solid organ transplantation

Study	Study characteristics	CMV disease[a]	CMV infection[a]	All-cause mortality[a]	Other effects
Antiviral prophylaxis					
Hodson et al.[46]	RCT of ganciclovir, aciclovir and valaciclovir prophylaxis vs placebo or no treatment	0.42 (0.34, 0.52) 19 trials 1981 patients	0.61 (0.48, 0.77) 17 trials 1786 patients	0.63 (0.43, 0.92) 17 trials 1838 patients	Reduction in HSV and VZV infections, bacterial and protozoal infections
Kalil et al.[47]	Prophylaxis vs placebo or no treatment	0.20 (0.13, 0.31) 11 trials 1582 patients	NA	0.62 (0.40, 0.96) 7 trials 1338 patients	Reduction in allograft rejection (RR: 0.74; 95% CI 0.59, 0.94)
Small et al.[49]	Ganciclovir prophylaxis vs placebo or no treatment	0.34 (0.24, 0.48) 8 trials 930 patients	NA	0.99 (0.68, 1.43) 12 trials 1322 patients	No significant reduction in rejection (RR: 0.90; 95% CI 0.79, 1.01)
Hodson et al.[50]	RCT comparing IgG with placebo or no treatment	0.80 (0.61, 1.05) 16 trials 770 patients	0.94 (0.80, 1.10) 14 trials 775 patients	0.57 (0.32, 1.03) 8 trials 502 patients	IgG reduced CMV-related deaths (RR: 0.33; 95% CI 0.14, 0.80) No differences in the risk of CMV disease, CMV infection and all-cause mortality between antiviral drug combined with IgG and antiviral medication alone
Pre-emptive therapy					
Hodson et al.[46]	RCT of pre-emptive therapy vs placebo or no treatment	0.29 (0.11, 0.80) 6 trials 288 patients	NA	1.23 (0.35, 4.30) 2 trials 176 patients	No significant effect of acute rejection (RR: 1.06; 95% CI 0.64, 1.76)
Kalil et al.[47]	Pre-emptive therapy vs placebo or no treatment	0.28 (0.11, 0.69) 6 trials 398 patients	NA	0.94 (0.32, 2.76) 3 trials 253 patients	Reduction in allograft rejection (RR: 0.47; 95% CI 0.24, 0.91)
Small et al.[49]	Pre-emptive therapy with ganciclovir	0.30 (0.15, 0.60) 9 trials 457 patients	NA	0.94 (0.43, 2.07) 4 trials 208 patients	No significant reduction in rejection (RR: 0.54; 95% CI 0.29, 1.01)

a Data presented as relative risk (95% confidence intervals), number of trials included and number of patients.

HSV = herpes simplex virus; **IgG** = immunoglobulin G; **NA** = not assessed; **RR** = relative risk; **VZV** = varicella-zoster virus.

Table IV. Prevention of cytomegalovirus (CMV) disease in solid organ transplant recipients[a]

Type of transplant	Modifying circumstance	Primary (preferred) strategy and agent(s)	Alternative strategy and agent(s)	Comments
Kidney, liver, pancreas and heart	D+/R–	Antiviral prophylaxis is preferred: valganciclovir 900 mg PO od × 3–6 months	Antiviral prophylaxis: ganciclovir 1 g PO tid Valaciclovir 2 g PO qid (for kidney recipients only) Pre-emptive therapy: valganciclovir 900 mg PO bid or ganciclovir 5 mg/kg IV every 12 hours for positive CMV PCR or antigenaemia test (duration of treatment is guided by CMV surveillance)	Valganciclovir is not US FDA-approved for liver transplant recipients but it is still the most commonly used drug for prophylaxis Late-onset CMV disease is the major complication of prophylaxis; occurring in 10–30% of D+/R– patients Six months of valganciclovir prophylaxis has been studied only in kidney recipients Recurrent CMV viraemia may occur with pre-emptive therapy, requiring repeated treatment Some centres add CMV hyperimmune globulin for heart recipients
	R+	Antiviral prophylaxis: valganciclovir 900 mg PO od × 3 months	Ganciclovir 1 g PO tid Valaciclovir 2 g PO qid (for kidney recipients only)	Low risk of late-onset CMV disease when compared with D+/R– patients Leukopenia and neutropenia are common adverse effects of ganciclovir Neurological toxicity occurs with high-dose valaciclovir
		Pre-emptive therapy: valganciclovir 900 mg PO bid for positive CMV PCR or antigenaemia (duration is guided by repeat CMV surveillance)	Ganciclovir 5 mg/kg IV every 12 hours for positive CMV PCR or antigenaemia test (duration guided by repeat CMV surveillance)	Oral ganciclovir and valaciclovir should not be used for pre-emptive therapy Foscarnet and cidofovir are not generally recommended for pre-emptive therapy because of high risk of toxicities
Lung (and heart-lung)	D+/R–	Antiviral prophylaxis is preferred: valganciclovir 900 mg PO od for at least 6 months	Antiviral prophylaxis: ganciclovir 5 mg/kg IV every 12 hours	Oral ganciclovir is not recommended because of poor absorption and the risk of drug resistance development Some centres start with IV ganciclovir and then transition to valganciclovir Some centres add unselected or CMV hyperimmune globulin
	R+	Antiviral prophylaxis is preferred: valganciclovir 900 mg PO od × 3–6 months	Antiviral prophylaxis is preferred: ganciclovir 5 mg/kg IV every 12 hours or ganciclovir 1 g PO tid Pre-emptive therapy: valganciclovir 900 mg PO bid or ganciclovir	CMV R+ lung transplant recipients are considered as high risk, and antiviral prophylaxis is preferred Oral ganciclovir is generally not preferred because of poor absorption and the risk of drug resistance development

Continued next page

Table IV. Contd

Type of transplant	Modifying circumstance	Primary (preferred) strategy and agent(s)	Alternative strategy and agent(s)	Comments
			5 mg/kg IV every 12 hours for positive CMV PCR or antigenaemia test (duration guided by repeated CMV surveillance)	
Intestinal	D+/R–	Antiviral prophylaxis is preferred: ganciclovir 5 mg/kg IV every 12 hours for 3–6 months		Pharmacokinetics of oral valganciclovir are not defined in intestinal transplant recipients Some centres add unselected or CMV hyperimmune globulin
	R+	Antiviral prophylaxis is preferred: ganciclovir 5 mg/kg IV every 12 hours for 3–6 months		CMV R+ intestinal transplant recipients are considered as high risk and antiviral prophylaxis is preferred

a Drug doses should be adjusted based on creatinine clearance.

bid = twice daily; **D+/R–** = donor positive/recipient negative; **IV** = intravenous; **od** = once daily; **PCR** = polymerase chain reaction; **PO** = by mouth; **qid** = four times daily; **R+** = recipient positive; **tid** = three times daily.

ganciclovir (1 g three times daily) in a large, randomized, controlled, multicentre, clinical trial that enrolled kidney, liver, pancreas and heart transplant recipients.[52] The study did not include lung and intestinal transplant recipients. In this study, both oral ganciclovir and valganciclovir had comparable efficacy in preventing CMV disease in CMV D+/R– SOT recipients.[52] However, in a subgroup analysis, liver transplant recipients who received valganciclovir developed more episodes of late-onset tissue-invasive CMV disease when compared with those who received oral ganciclovir prophylaxis.[52] Because of this finding, valganciclovir was not approved by the US FDA for prevention of CMV disease in liver transplant recipients. Nonetheless, despite these results and the FDA call for caution, valganciclovir is widely used for prophylaxis in liver transplant recipients. In a smaller study of lung transplant recipients, valganciclovir prophylaxis was also found to be as efficacious as oral ganciclovir in preventing CMV disease.[53]

There is currently no recommendation on the use of immunoglobulins for CMV prevention after SOT.[50] Compared with placebo or no treatment, the incidences of CMV disease, CMV infection and all-cause mortality among patients who received immunoglobulins were not significantly reduced.[50] Moreover, the addition of immunoglobulins to antiviral drugs did not provide added benefit, in terms of further reduction in the incidence of CMV disease, when compared with SOT recipients who were receiving antiviral medication only. Interestingly, immunoglobulin use was associated with lower death rate from CMV-related causes.[50]

Until recently, the investigational drug maribavir, a benzimidazole riboside that inhibits viral DNA assembly and the egress of viral capsids from the cell, was being developed for the prevention of CMV disease in allogeneic haematopoietic stem cell (HSCT) and liver transplant recipients. *In vitro* studies have shown activity against CMV resistant to ganciclovir, cidofovir or foscarnet.[54] Data from a phase II clinical trial investigating the use of maribavir for prophylaxis in allogeneic HSCT recipients were encouraging, with lower rates of CMV infection in patients

who received maribavir when compared with placebo.[55] However, a recent phase III study of maribavir in allogeneic HSCT recipients did not confirm these observations and found that it was not significantly better than the placebo comparator. The incidence of CMV disease within 6 months was 4.4% in the maribavir group and 4.8% in the placebo group ($p = 0.79$). The rates of CMV DNAaemia were 30.4% in the placebo group and 27.8% in the maribavir group. There were no significant differences between maribavir and placebo for either the primary endpoint (CMV disease within 6 months) or for any of the secondary endpoints (i.e. rate of anti-CMV treatment within 6 months, incidence of graft-versus-host disease, mortality and CMV disease-free survival). As a result of the discouraging data from the latter study, the phase III trial comparing maribavir and oral ganciclovir prophylaxis in CMV D+/R− liver transplant recipients was discontinued. In the interim analysis of this prematurely halted liver transplant trial, patients who were receiving maribavir reportedly had higher rates of CMV infection than those who received oral ganciclovir prophylaxis.

3.1.2 Duration of Prophylaxis

Virtually no cases of breakthrough CMV disease occur during the time of antiviral prophylaxis. However, almost all cases of CMV disease occur after the cessation of antiviral prophylaxis – hence the term delayed-onset (or late-onset) CMV disease.[3,5,6] The 1-year incidence of delayed-onset CMV disease among CMV D+/R− liver, heart, kidney and pancreas recipients who received prophylaxis for 3 months is 29%.[3,5,6,13] The median time-to-CMV disease is 55 days after the end of prophylaxis, hence the onset of disease in most cases occurs during the first 3 months after cessation of prophylaxis.[3,5,6] While one study found no significant association between late-onset CMV disease and transplant outcomes,[7] two studies observed that delayed-onset CMV disease was associated with poor patient and graft survival after kidney and liver transplantation.[2,3] Therefore, research efforts have been focused on finding better preventive strategies. In this regard, one study tested the use of pre-emptive therapy following completion of prophylaxis, but found the strategy to be complicated logistically and this resulted in modest compliance with CMV surveillance.[56] Another potential strategy consists of extending the duration of antiviral prophylaxis, mainly in high-risk CMV D+/R− SOT patients. The potential drawbacks to this strategy would be an increased incidence of drug resistance and drug toxicity, and higher drug costs. A small single-centre study of 68 CMV D+/R− kidney recipients demonstrated a lower incidence of CMV disease among those who received 24 weeks compared with 12 weeks of oral ganciclovir prophylaxis (7% vs 31%, respectively).[57]

To further investigate this approach, an international, randomized, prospective, double-blind trial (IMPACT [Improved Protection Against Cytomegalovirus in Transplant]) was conducted. In this study, 318 CMV D+/R− kidney transplant recipients received prophylaxis with either valganciclovir 900 mg once daily (or equivalent dose after adjusting for the renal function) for 100 days followed by placebo until 200 days after transplantation or valganciclovir 900 mg once daily for 200 days after transplantation. CMV disease, allograft rejection and loss, and safety were analysed up to 12 months after transplant. Confirmed CMV disease, defined as CMV syndrome or tissue-invasive disease, developed in 36.8% versus 16.1% of patients in the 100-day prophylaxis group versus the 200-day prophylaxis group, respectively ($p < 0.0001$). The rates of acute rejection (17.2% vs 11%; $p = 0.11$) and allograft loss (1.8% vs 1.9%; $p = 0.9$) were low and similar between the 100-day and 200-day prophylaxis groups. There was no significant difference in overall tolerability, and the incidence and grading of haematological parameters (neutrophils, haemoglobin and platelets) were similar between groups. The authors concluded that, overall, 200 days of once-daily oral valganciclovir prophylaxis provides a significant reduction in the incidence of CMV disease compared with 100 days of prophylaxis in high-risk kidney transplant recipients, as assessed 12 months after transplant, without adversely impacting on safety.[58] As a result of this study, the new American

Society of Transplantation guidelines now recommend 3–6 months of antiviral prophylaxis for high-risk CMV D+/R− SOT recipients.[48]

Recently, a small single-centre study compared the use of delayed primary CMV prophylaxis (wherein antiviral prophylaxis was not started until 2 weeks after SOT) and conventional prophylaxis (wherein antiviral prophylaxis is started immediately after transplantation). The underlying hypothesis is that delayed prophylaxis will allow transient exposure of the immune system to CMV, which would lead to the development of adequate CMV-specific immunity while the virus is suppressed. Interestingly, CMV disease occurred in 7 of 26 patients (27%) receiving conventional prophylaxis compared with only 1 of 18 patients (5.5%) receiving delayed prophylaxis ($p = 0.07$). Furthermore, five patients (19%) receiving conventional prophylaxis developed CMV colitis, while none of the patients receiving delayed prophylaxis developed tissue-invasive disease ($p = 0.048$).[59] The observations in this small study are interesting enough that this approach should be investigated in a larger controlled clinical trial.

3.2 Pre-Emptive Therapy

This strategy consists of weekly laboratory monitoring to detect CMV in the blood using pp65 antigen assay or polymerase chain reaction (PCR) prior to the onset of symptoms. Once CMV is detected, pre-emptive therapy is initiated to prevent the progression of asymptomatic infection to clinical disease.[4,60,61] The potential advantage of pre-emptive therapy is the exposure of the host's immune system to CMV, thereby allowing earlier CMV-specific T-cell reconstitution, which could potentially explain the very low rates of late-onset CMV disease with this approach. However, the success of pre-emptive therapy is highly dependent on the consistency of collecting weekly specimens and the pre-dictive value of the assay used. Most transplant centres currently use CMV PCR, which is known to have very high sensitivity and specificity, allowing early detection of CMV in the preclinical phase. Furthermore, the ability to provide patients with CMV PCR kits makes the collection of specimens through local laboratories possible and, therefore, improves patient compliance. Indeed, in one study, the major limitation compromising the success of this strategy was compliance;[56] 41% of patients who developed CMV disease did not submit at least one required sample prior to CMV disease.[56] Among CMV D+/R− patients, pre-emptive therapy is further undermined by the rapid replication of the virus in these high-risk patients[62] and it may result in clinical disease prior to CMV detection by a weekly PCR assay.[60,61] In our clinical experience, one out of four CMV D+/R− liver recipients develop CMV disease while being monitored with CMV weekly PCR.[60,61] Accordingly, current guidelines do not recommend pre-emptive therapy for the prevention of CMV disease in high-risk CMV D+/R− SOT recipients.

There have been multiple studies that assessed the efficacy of pre-emptive therapy in preventing CMV infection and disease; the results of those studies were collectively analysed in three meta-analyses (table III). In all three studies, the average reduction in incidence of CMV disease was 70%. In addition, pre-emptive therapy was as effective as primary prophylaxis in preventing CMV disease[46,47,49] and the costs are similar.[63] Acute rejection was reduced in one study.[47] However, pre-emptive antiviral therapy did not reduce the incidence of all-cause mortality.[46,47,49]

Currently, valganciclovir is the drug of choice for the pre-emptive treatment of patients with asymptomatic detectable CMV viraemia. In one study, pre-emptive valganciclovir therapy had a primary efficacy of 79% in preventing progression to CMV disease among kidney transplant recipients.[64] This was also demonstrated in another study of CMV-seropositive SOT recipients with detectable CMV antigenaemia.[65]

4. Treatment of CMV Disease in SOT Recipients

Transplant patients presenting with CMV disease should be carefully evaluated to define the extent of their disease. While most patients present with CMV syndrome, some present with

severe and possibly life-threatening tissue-invasive disease. Since the severity of disease and clinical presentation is influenced by the net state of immunosuppression, it is generally recommended that, in addition to antiviral drug therapy, the doses of immunosuppressive medications should be reduced to the lowest safe dose in all patients.

Intravenous ganciclovir remains the drug of choice for the treatment of CMV disease in transplant recipients,[48] especially those with severe infection. In contrast, oral ganciclovir should not be used for the treatment of CMV disease because of the limited absorption and poor bioavailability. More recently, the introduction of valganciclovir has allowed for the oral treatment of CMV disease in SOT recipients.[4] In a recent clinical study, CMV decay was similar between intravenous ganciclovir and valganciclovir.[66,67]

Indeed, valganciclovir was recently shown to be as effective as intravenous ganciclovir in the treatment of mild-to-moderate (i.e. non-severe) CMV disease. A total of 321 SOT recipients with CMV disease were randomized to receive induction therapy with valganciclovir or intravenous ganciclovir for 21 days followed by valganciclovir maintenance therapy for 4 weeks. The proportions of viral eradication at day 21 and day 49 were comparable between the intravenous ganciclovir and valganciclovir groups. Similarly, resolution of clinical symptomatology was not significantly different between valganciclovir and intravenous ganciclovir. The overall time to viral eradication was 21 days with valganciclovir and 19 days with intravenous ganciclovir. The calculated viral decay was 11.5 days with valganciclovir and 10.4 days with intravenous ganciclovir.[68] It is worth emphasizing that the patients enrolled in this study were mostly CMV-seropositive kidney recipients and patients with severe CMV disease were excluded. This pivotal trial supports the use of valganciclovir for the oral treatment of CMV disease, at least in SOT patients with mild-to-moderate disease. During long-term follow-up at 1 year, clinical recurrence (beyond day 49) was 14.8% among valganciclovir recipients and 15.5% among intravenous ganciclovir recipients ($p = 0.89$).[69] The rate of virological recurrence was also not significantly different between the two groups (30.9% in the valganciclovir group and 29.1% in the intravenous ganciclovir group; $p = 0.77$).

The duration of treatment for CMV disease should be individualized, and guided by virological and clinical surveillance. Undetectable viraemia should be achieved prior to discontinuation of therapy in order to reduce the risk of clinical relapse. Previous studies have shown that persistent viraemia at the end of therapy is associated with a higher risk of disease relapse.[70,71] At least two CMV PCR assays performed 1 week apart should be negative prior to discontinuation of treatment. However, occasionally tissue-invasive disease is not accompanied by viraemia (also known as 'compartmentalized disease'), which limits the ability to use serum PCR assays to guide therapy.

CMV gastrointestinal disease is the most common form of CMV tissue-invasive disease.[72] Clinical relapse following treatment is not uncommon, especially in high-risk patients. A recent study investigated the clinical predictors of relapse among CMV D+/R– SOT recipients with biopsy-proven CMV gastrointestinal disease.[25] CMV relapse, which occurred in 27% of patients, was significantly associated with extensive disease defined as upper and lower gastrointestinal tract involvement. CMV seroconversion, viral load, treatment duration, maintenance therapy and endoscopic evidence of resolution of gastrointestinal disease were not associated with lower risk of CMV relapse.[25] According to those results, end-of-treatment colonoscopy or upper endoscopy should not be routinely recommended to document clearance of gastrointestinal CMV disease prior to discontinuation of therapy.

4.1 Treatment of Ganciclovir-Resistant CMV Disease

In the current era, ganciclovir-resistant CMV has become an increasingly significant clinical challenge.[73] Ganciclovir resistance is more likely to develop among CMV D+/R– patients, lung and pancreas recipients, and in the context of high-level viral replication, potent immunosuppressive therapy and suboptimal ganciclovir levels.[4,74,75]

The incidence of ganciclovir-resistant CMV with oral ganciclovir prophylaxis ranged from 0% (among liver transplant recipients) to 13% (among kidney-pancreas transplant recipients).[74] While the incidence appears to be lower with valganciclovir prophylaxis, which was estimated at <4%, this may rise with prolonged use of the drug, especially with suboptimal dosing in high-risk CMV D+/R− recipients. Most cases of ganciclovir-resistance are attributed to a *UL97* mutation.[74,76-78] *UL97* is the gene that encodes for a viral kinase that initially phosphorylates ganciclovir into its active form. Hence, a mutation in the *UL97* gene would keep the ganciclovir in its inactive non-phosphorylated form. A mutation in the *UL54* gene, which encodes for the CMV DNA polymerase, the target for all available systemic anti-CMV drugs, could occur independently or following *UL97* mutation, and it could lead to cidofovir and/or foscarnet resistance or cross-resistance among ganciclovir, foscarnet and cidofovir.

The treatment options for ganciclovir-resistant CMV are limited. In addition, most antiviral drugs available to treat ganciclovir-resistant CMV have serious adverse effects, which could explain the significant morbidity and mortality associated with this disease. Among 225 CMV D+/R− SOT recipients who received 3 months of valganciclovir prophylaxis, CMV disease occurred in 65 patients (29%), including four (8%) that were due to drug-resistant CMV.[79] Those four patients were treated with foscarnet or cidofovir, and two of them developed drug-induced nephrotoxicity and allograft loss.[79] Other potential therapeutic agents for multi-drug-resistant CMV include immunoglobulins, leflunomide[80] and artesunate,[81,82] although data supporting their use remains anecdotal.[83] Hence, there is a need to identify novel agents and strategies for the management of CMV infection and disease. Currently, artesunate is undergoing a phase III trial in HSCT recipients. Despite the failure of maribavir as a primary agent for the prevention of wild-type CMV disease, it could still be considered an option for treatment of multi-drug-resistant CMV as it has a completely different mechanism of action compared with currently available and approved agents. However, its clinical development is currently on hold.

Other compounds that are currently undergoing investigation for use in the management of CMV infection are CMX-001 and the immunosuppressive drugs sirolimus and everolimus. CMX-001, an ester formulation of cidofovir, has the same mechanism of action as the parent compound (and may not offer a novel strategy) but it is reported to have a lesser toxicity profile. Sirolimus and everolimus have been observed, anecdotally and in clinical immunosuppression trials, to be associated with a lower incidence of CMV disease.[84,85] Hence, everolimus, a macrocyclic immunosuppressant that blocks growth factor-driven transduction signals, is now undergoing a phase II trial in kidney transplant recipients as an add-on treatment.

There are several CMV vaccine candidates (CMV gB vaccine, VCL-CB01, CMV pp65 peptide, ALVAC-CMV, VCL-CT02) that are undergoing early-phase clinical trials in healthy and transplant recipients.[86] Previous data using the attenuated CMV vaccine (Towne strain) showed that CMV infection was not prevented in vaccinated transplant candidates but the severity of CMV disease was markedly reduced.[87] With this knowledge, the major aim of CMV vaccination trials should therefore be focused on inducing CMV-specific immunity in order to reduce not only the clinical consequences of primary CMV infection, but also the severity of CMV re-infections and reactivation in at-risk individuals.[86] The transfer of CMV-specific T cells through adoptive immunotherapy has also been evaluated in pilot clinical studies for the prevention and treatment of CMV disease after transplantation, with promising results.[88,89] This is now being investigated in controlled clinical trials to assess its safety and efficacy for prevention[90] and treatment of persistent or refractory CMV infections[91] after transplantation.[86]

5. Conclusions

Despite remarkable advances in its prevention and treatment, CMV remains a common disease that negatively influences the outcome of SOT

recipients. In addition to viral factors and pharmacological immunosuppression, the roles of innate and adaptive immune deficiencies are now being recognized in its pathogenesis. Such novel findings should provide additional avenues and opportunities for improving our management strategies. Prevention of CMV with antiviral prophylaxis and pre-emptive therapy are both effective. However, delayed-onset primary CMV disease is a common consequence of antiviral prophylaxis. Extending the duration of prophylaxis to 6 months may further reduce the incidence of CMV disease. Intravenous ganciclovir and oral valganciclovir are equally effective treatment of CMV disease, although intravenous ganciclovir should remain the drug of choice for patients with severe disease and those with high viral loads. Ganciclovir-resistant CMV is increasingly being recognized in the SOT population and, while it remains rare, the morbidity associated with it is enormous; as antiviral drug use becomes widespread, we anticipate an increase in the incidence of drug-resistant CMV. While judicious use of antiviral drugs is likely to curtail drug-resistant CMV, we also encourage the development of novel drugs in order to combat this emerging challenge.

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References

1. Slifkin M, Doron S, Snydman DR. Viral prophylaxis in organ transplant patients. Drugs 2004; 64 (24): 2763-92
2. Limaye AP, Bakthavatsalam R, Kim HW, et al. Impact of cytomegalovirus in organ transplant recipients in the era of antiviral prophylaxis. Transplantation 2006; 81 (12): 1645-52
3. Arthurs SK, Eid AJ, Pedersen RA, et al. Delayed-onset primary cytomegalovirus disease and the risk of allograft failure and mortality after kidney transplantation. Clin Infect Dis 2008; 46 (6): 840-6
4. Razonable RR, Paya CV. Herpes virus infections in transplant recipients: current challenges in the clinical management of cytomegalovirus and Epstein-Barr virus infections. Herpes 2003; 10 (3): 60-5
5. Arthurs SK, Eid AJ, Pedersen RA, et al. Delayed-onset primary cytomegalovirus disease after liver transplantation. Liver Transpl 2007; 13 (12): 1703-9
6. Kijpittayarit-Arthurs S, Eid AJ, Kremers WK, et al. Clinical features and outcomes of delayed-onset primary cytomegalovirus disease in cardiac transplant recipients. J Heart Lung Transplant 2007; 26 (10): 1019-24
7. Scemla A, Kreis H, Legendre C. Less pessimistic long-term results for patients with cytomegalovirus disease. Clin Infect Dis 2008; 47 (10): 1360-1; author reply 1361-2
8. Kumar D, Chernenko S, Moussa G, et al. Cell-mediated immunity to predict cytomegalovirus disease in high-risk solid organ transplant recipients. Am J Transplant 2009; 9 (5): 1214-22
9. Peleg AY, Husain S, Kwak EJ, et al. Opportunistic infections in 547 organ transplant recipients receiving alemtuzumab, a humanized monoclonal CD-52 antibody. Clin Infect Dis 2007; 44 (2): 204-12
10. Zeevi A, Husain S, Spichty KJ, et al. Recovery of functional memory T cells in lung transplant recipients following induction therapy with alemtuzumab. Am J Transplant 2007; 7 (2): 471-5
11. Hoppe L, Marroni CA, Bressane R, et al. Risk factors associated with cytomegalovirus infection in orthotopic liver transplant patients. Transplant Proc 2006; 38 (6): 1922-3
12. Song AT, Abdala E, Bonazzi PR, et al. Does mycophenolate mofetil increase the risk of cytomegalovirus infection in solid organ transplant recipients? A mini-review. Braz J Infect Dis 2006; 10 (2): 132-8
13. Razonable RR, Rivero A, Rodriguez A, et al. Allograft rejection predicts the occurrence of late-onset cytomegalovirus (CMV) disease among CMV-mismatched solid organ transplant patients receiving prophylaxis with oral ganciclovir. J Infect Dis 2001; 184 (11): 1461-4
14. Reusser P, Cathomas G, Attenhofer R, et al. Cytomegalovirus (CMV)-specific T cell immunity after renal transplantation mediates protection from CMV disease by limiting the systemic virus load. J Infect Dis 1999; 180 (2): 247-53
15. Sester M, Sester U, Gartner B, et al. Levels of virus-specific CD4 T cells correlate with cytomegalovirus control and predict virus-induced disease after renal transplantation. Transplantation 2001; 71 (9): 1287-94
16. Tu W, Potena L, Stepick-Biek P, et al. T-cell immunity to subclinical cytomegalovirus infection reduces cardiac allograft disease. Circulation 2006; 114 (15): 1608-15
17. Gerna G, Lilleri D, Fornara C, et al. Monitoring of human cytomegalovirus-specific CD4 and CD8 T-cell immunity in patients receiving solid organ transplantation. Am J Transplant 2006; 6 (10): 2356-64
18. Eid AJ, Brown RA, Arthurs SK, et al. A prospective longitudinal analysis of cytomegalovirus (CMV)-specific CD4+ and CD8+ T cells in kidney allograft recipients at risk of CMV infection. Transpl Int. Epub 2009 Nov 27
19. La Rosa C, Limaye AP, Krishnan A, et al. Longitudinal assessment of cytomegalovirus (CMV)-specific immune responses in liver transplant recipients at high risk for late CMV disease. J Infect Dis 2007; 195 (5): 633-44
20. Kijpittayarit S, Eid AJ, Brown RA, et al. Relationship between Toll-like receptor 2 polymorphism and cytomegalovirus

disease after liver transplantation. Clin Infect Dis 2007; 44 (10): 1315-20

21. Ducloux D, Deschamps M, Yannaraki M, et al. Relevance of Toll-like receptor-4 polymorphisms in renal transplantation. Kidney Int 2005; 67 (6): 2454-61
22. Cervera C, Lozano F, Saval N, et al. The influence of innate immunity gene receptors polymorphisms in renal transplant infections. Transplantation 2007; 83 (11): 1493-500
23. Manuel O, Pascual M, Trendelenburg M, et al. Association between mannose-binding lectin deficiency and cytomegalovirus infection after kidney transplantation. Transplantation 2007; 83 (3): 359-62
24. Loeffler J, Steffens M, Arlt EM, et al. Polymorphisms in the genes encoding chemokine receptor 5, interleukin-10, and monocyte chemoattractant protein 1 contribute to cytomegalovirus reactivation and disease after allogeneic stem cell transplantation. J Clin Microbiol 2006; 44 (5): 1847-50
25. Eid AJ, Arthurs SK, Deziel PJ, et al. Clinical predictors of relapse after treatment of primary gastrointestinal cytomegalovirus disease in solid organ transplant recipients. Am J Transplant 2010 Jan; 10 (1): 157-61
26. Eid AJ, Bakri SJ, Kijpittayarit S, et al. Clinical features and outcomes of cytomegalovirus retinitis after transplantation. Transpl Infect Dis 2008 Feb; 10 (1): 13-8
27. Razonable RR, Brown RA, Humar A, et al. A longitudinal molecular surveillance study of human polyomavirus viremia in heart, kidney, liver, and pancreas transplant patients. J Infect Dis 2005; 192 (8): 1349-54
28. Razonable RR, Paya CV. The impact of human herpesvirus-6 and -7 infection on the outcome of liver transplantation. Liver Transpl 2002; 8 (8): 651-8
29. Razonable RR, Rivero A, Brown RA, et al. Detection of simultaneous beta-herpesvirus infections in clinical syndromes due to defined cytomegalovirus infection. Clin Transplant 2003; 17 (2): 114-20
30. Peleg AY, Husain S, Qureshi ZA, et al. Risk factors, clinical characteristics, and outcome of Nocardia infection in organ transplant recipients: a matched case-control study. Clin Infect Dis 2007; 44 (10): 1307-14
31. Husni RN, Gordon SM, Longworth DL, et al. Cytomegalovirus infection is a risk factor for invasive aspergillosis in lung transplant recipients. Clin Infect Dis 1998; 26 (3): 753-5
32. Razonable RR, Burak KW, van Cruijsen H, et al. The pathogenesis of hepatitis C virus is influenced by cytomegalovirus. Clin Infect Dis 2002; 35 (8): 974-81
33. Nebbia G, Mattes FM, Cholongitas E, et al. Exploring the bidirectional interactions between human cytomegalovirus and hepatitis C virus replication after liver transplantation. Liver Transpl 2007; 13 (1): 130-5
34. Fearon WF, Potena L, Hirohata A, et al. Changes in coronary arterial dimensions early after cardiac transplantation. Transplantation 2007; 83 (6): 700-5
35. Hussain T, Burch M, Fenton MJ, et al. Positive pretransplantation cytomegalovirus serology is a risk factor for cardiac allograft vasculopathy in children. Circulation 2007; 115 (13): 1798-805
36. Ozdemir FN, Akgul A, Altunoglu A, et al. The association between cytomegalovirus infection and atherosclerotic events in renal transplant recipients. Transplant Proc 2007; 39 (4): 990-2
37. Thomas LD, Milstone AP, Miller GG, et al. Long-term outcomes of cytomegalovirus infection and disease after lung or heart-lung transplantation with a delayed ganciclovir regimen. Clin Transplant 2009; 23 (4): 476-83
38. Rodrigo E, Fernandez-Fresnedo G, Valero R, et al. New-onset diabetes after kidney transplantation: risk factors. J Am Soc Nephrol 2006; 17 (12 Suppl. 3): S291-5
39. Hartmann A, Sagedal S, Hjelmesaeth J. The natural course of cytomegalovirus infection and disease in renal transplant recipients. Transplantation 2006; 82 (2 Suppl.): S15-7
40. Couzi L, Levaillant Y, Jamai A, et al. Cytomegalovirus-induced gammadelta T cells associate with reduced cancer risk after kidney transplantation. J Am Soc Nephrol 2010 Jan; 21 (1): 181-8
41. Sagedal S, Rollag H, Hartmann A. Cytomegalovirus infection in renal transplant recipients is associated with impaired survival irrespective of expected mortality risk. Clin Transplant 2007; 21 (3): 309-13
42. Helantera I, Koskinen P, Finne P, et al. Persistent cytomegalovirus infection in kidney allografts is associated with inferior graft function and survival. Transpl Int 2006; 19 (11): 893-900
43. Potena L, Holweg CT, Chin C, et al. Acute rejection and cardiac allograft vascular disease is reduced by suppression of subclinical cytomegalovirus infection. Transplantation 2006; 82 (3): 398-405
44. Hellinger WC, Bonatti H, Machicao VI, et al. Effect of antiviral chemoprophylaxis on adverse clinical outcomes associated with cytomegalovirus after liver transplantation. Mayo Clin Proc 2006; 81 (8): 1029-33
45. Manuel O, Kumar D, Moussa G, et al. Lack of association between beta-herpesvirus infection and bronchiolitis obliterans syndrome in lung transplant recipients in the era of antiviral prophylaxis. Transplantation 2009; 87 (5): 719-25
46. Hodson EM, Jones CA, Webster AC, et al. Antiviral medications to prevent cytomegalovirus disease and early death in recipients of solid-organ transplants: a systematic review of randomised controlled trials. Lancet 2005; 365 (9477): 2105-15
47. Kalil AC, Levitsky J, Lyden E, et al. Meta-analysis: the efficacy of strategies to prevent organ disease by cytomegalovirus in solid organ transplant recipients. Ann Intern Med 2005; 143 (12): 870-80
48. Humar A, Snydman D. Cytomegalovirus in solid organ transplant recipients. Am J Transplant 2009; 9 Suppl. 4: S78-86
49. Small LN, Lau J, Snydman DR. Preventing post-organ transplantation cytomegalovirus disease with ganciclovir: a meta-analysis comparing prophylactic and preemptive therapies. Clin Infect Dis 2006; 43 (7): 869-80
50. Hodson EM, Jones CA, Strippoli GF, et al. Immunoglobulins, vaccines or interferon for preventing cytomegalovirus disease in solid organ transplant recipients. Cochrane Database Syst Rev 2007; (2): CD005129
51. Hostetler KY. Alkoxyalkyl prodrugs of acyclic nucleoside phosphonates enhance oral antiviral activity and reduce toxicity: current state of the art. Antiviral Res 2009 May; 82 (2): A84-98
52. Paya C, Humar A, Dominguez E, et al. Efficacy and safety of valganciclovir vs. oral ganciclovir for prevention of

cytomegalovirus disease in solid organ transplant recipients. Am J Transplant 2004; 4 (4): 611-20

53. Humar A, Kumar D, Preiksaitis J, et al. A trial of valganciclovir prophylaxis for cytomegalovirus prevention in lung transplant recipients. Am J Transplant 2005; 5 (6): 1462-8
54. Drew WL, Miner RC, Marousek GI, et al. Maribavir sensitivity of cytomegalovirus isolates resistant to ganciclovir, cidofovir or foscarnet. J Clin Virol 2006; 37 (2): 124-7
55. Winston DJ, Young JA, Pullarkat V, et al. Maribavir prophylaxis for prevention of cytomegalovirus infection in allogeneic stem cell transplant recipients: a multicenter, randomized, double-blind, placebo-controlled, dose-ranging study. Blood 2008; 111 (11): 5403-10
56. Walker JK, Scholz LM, Scheetz MH, et al. Leukopenia complicates cytomegalovirus prevention after renal transplantation with alemtuzumab induction. Transplantation 2007; 83 (7): 874-82
57. Doyle AM, Warburton KM, Goral S, et al. 24-week oral ganciclovir prophylaxis in kidney recipients is associated with reduced symptomatic cytomegalovirus disease compared to a 12-week course. Transplantation 2006; 81 (8): 1106-11
58. Humar A, Lebranchu Y, Vincenti F, et al. The efficacy and safety of 200 days valganciclovir cytomegalovirus prophylaxis in high-risk kidney transplant recipients. Am J Transplant. In Press
59. San Juan R, Yebra M, Lumbreras C, et al. A new strategy of delayed long-term prophylaxis could prevent cytomegalovirus disease in (D+/R–) solid organ transplant recipients. Clin Transplant 2009; 23 (5): 666-71
60. Razonable RR, van Cruijsen H, Brown RA, et al. Dynamics of cytomegalovirus replication during preemptive therapy with oral ganciclovir. J Infect Dis 2003; 187 (11): 1801-8
61. Paya CV, Wilson JA, Espy MJ, et al. Preemptive use of oral ganciclovir to prevent cytomegalovirus infection in liver transplant patients: a randomized, placebo-controlled trial. J Infect Dis 2002; 185 (7): 854-60
62. Emery VC, Hassan-Walker AF, Burroughs AK, et al. Human cytomegalovirus (HCMV) replication dynamics in HCMV-naive and -experienced immunocompromised hosts. J Infect Dis 2002; 185 (12): 1723-8
63. Khoury JA, Storch GA, Bohl DL, et al. Prophylactic versus preemptive oral valganciclovir for the management of cytomegalovirus infection in adult renal transplant recipients. Am J Transplant 2006; 6 (9): 2134-43
64. Lopau K, Greser A, Wanner C. Efficacy and safety of preemptive anti-CMV therapy with valganciclovir after kidney transplantation. Clin Transplant 2007; 21 (1): 80-5
65. Diaz-Pedroche C, Lumbreras C, San Juan R, et al. Valganciclovir preemptive therapy for the prevention of cytomegalovirus disease in high-risk seropositive solid-organ transplant recipients. Transplantation 2006; 82 (1): 30-5
66. Mattes FM, Hainsworth EG, Hassan-Walker AF, et al. Kinetics of cytomegalovirus load decrease in solid-organ transplant recipients after preemptive therapy with valganciclovir. J Infect Dis 2005; 191 (1): 89-92
67. Humar A, Siegal D, Moussa G, et al. A prospective assessment of valganciclovir for the treatment of cytomegalovirus infection and disease in transplant recipients. J Infect Dis 2005; 192 (7): 1154-7
68. Asberg A, Humar A, Rollag H, et al. Oral valganciclovir is noninferior to intravenous ganciclovir for the treatment of cytomegalovirus disease in solid organ transplant recipients. Am J Transplant 2007; 7 (9): 2106-13
69. Asberg A, Humar A, Jardine AG, et al. Long-term outcomes of CMV disease treatment with valganciclovir versus IV ganciclovir in solid organ transplant recipients. Am J Transplant 2009; 9 (5): 1205-13
70. Humar A, Kumar D, Boivin G, et al. Cytomegalovirus (CMV) virus load kinetics to predict recurrent disease in solid-organ transplant patients with CMV disease. J Infect Dis 2002; 186 (6): 829-33
71. Sia IG, Wilson JA, Groettum CM, et al. Cytomegalovirus (CMV) DNA load predicts relapsing CMV infection after solid organ transplantation. J Infect Dis 2000; 181 (2): 717-20
72. Fica A, Cervera C, Perez N, et al. Immunohistochemically proven cytomegalovirus end-organ disease in solid organ transplant patients: clinical features and usefulness of conventional diagnostic tests. Transpl Infect Dis 2007; 9 (3): 203-10
73. Nogueira E, Ozaki KS, Tomiyama H, et al. The emergence of cytomegalovirus resistance to ganciclovir therapy in kidney transplant recipients. Int Immunopharmacol 2006; 6 (13-14): 2031-7
74. Limaye AP. Antiviral resistance in cytomegalovirus: an emerging problem in organ transplant recipients. Semin Respir Infect 2002; 17 (4): 265-73
75. Limaye AP, Raghu G, Koelle DM, et al. High incidence of ganciclovir-resistant cytomegalovirus infection among lung transplant recipients receiving preemptive therapy. J Infect Dis 2002; 185 (1): 20-7
76. Boivin G, Goyette N, Gilbert C, et al. Analysis of cytomegalovirus DNA polymerase (UL54) mutations in solid organ transplant patients receiving valganciclovir or ganciclovir prophylaxis. J Med Virol 2005; 77 (3): 425-9
77. Boivin G, Gilbert C, Gaudreau A, et al. Rate of emergence of cytomegalovirus (CMV) mutations in leukocytes of patients with acquired immunodeficiency syndrome who are receiving valganciclovir as induction and maintenance therapy for CMV retinitis. J Infect Dis 2001; 184 (12): 1598-602
78. Mendez JC, Sia IG, Tau KR, et al. Novel mutation in the CMV UL97 gene associated with resistance to ganciclovir therapy. Transplantation 1999; 67 (5): 755-7
79. Eid AJ, Arthurs SK, Deziel PJ, et al. Emergence of drug-resistant cytomegalovirus in the era of valganciclovir prophylaxis: therapeutic implications and outcomes. Clin Transplant 2008; 22 (2): 162-70
80. Battiwalla M, Paplham P, Almyroudis NG, et al. Leflunomide failure to control recurrent cytomegalovirus infection in the setting of renal failure after allogeneic stem cell transplantation. Transpl Infect Dis 2007; 9 (1): 28-32
81. Efferth T, Romero MR, Wolf DG, et al. The antiviral activities of artemisinin and artesunate. Clin Infect Dis 2008; 47 (6): 804-11
82. Shapira MY, Resnick IB, Chou S, et al. Artesunate as a potent antiviral agent in a patient with late drug-resistant cytomegalovirus infection after hematopoietic stem cell transplantation. Clin Infect Dis 2008; 46 (9): 1455-7
83. Levi ME, Mandava N, Chan LK, et al. Treatment of multidrug-resistant cytomegalovirus retinitis with systemically

administered leflunomide. Transpl Infect Dis 2006; 8 (1): 38-43

84. Hill JA, Hummel M, Starling RC, et al. A lower incidence of cytomegalovirus infection in de novo heart transplant recipients randomized to everolimus. Transplantation 2007; 84 (11): 1436-42
85. San Juan R, Aguado JM, Lumbreras C, et al. Impact of current transplantation management on the development of cytomegalovirus disease after renal transplantation. Clin Infect Dis 2008; 47 (7): 875-82
86. Razonable RR. Immune-based therapies for cytomegalovirus infection. Immunotherapy 2010; 2 (1): 117-30
87. Plotkin SA, Smiley ML, Friedman HM, et al. Towne-vaccine-induced prevention of cytomegalovirus disease after renal transplants. Lancet 1984; 1 (8376): 528-30
88. Cobbold M, Khan N, Pourgheysari B, et al. Adoptive transfer of cytomegalovirus-specific CTL to stem cell transplant patients after selection by HLA-peptide tetramers. J Exp Med 2005; 202 (3): 379-86
89. Bao L, Dunham K, Stamer M, et al. Expansion of cytomegalovirus pp65 and IE-1 specific cytotoxic T lymphocytes for cytomegalovirus-specific immunotherapy following allogeneic stem cell transplantation. Biol Blood Marrow Transplant 2008; 14 (10): 1156-62
90. Slavin S. Adoptive immunotherapy for CMV disease [ClinicalTrials.gov identifier NCT00159055]. US National Institutes of Health, ClinicalTrials.gov [online]. Available from URL: http://www.clinicaltrials.gov [Accessed 2010 Mar 23]
91. Lucas KG. Vaccine therapy in treating patients who have undergone a donor stem cell transplant and have cytomegalovirus infection that has not responded to therapy [ClinicalTrials.gov identifier NCT00509691]. US National Institutes of Health, ClinicalTrials.gov [online]. Available from URL: http://www.clinicaltrials.gov [Accessed 2010 Mar 23]

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