

Continuing Medical Education

CMV: Prevention, Diagnosis and Therapy

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Cytomegalovirus (CMV) is the most common infection after organ transplantation and has a major impact on morbidity, mortality and graft survival. Optimal prevention, diagnosis and treatment of active CMV infection enhance transplant outcomes, and are the focus of this section. Methods to prevent CMV include universal prophylaxis and preemptive therapy; each has its merits, and will be compared and contrasted. Diagnostics have improved substantially in recent years, both in type and quality, allowing for more accurate and savvy treatment; advances in diagnostics include the development of an international standard, which should allow comparison of results across different methodologies, and assays for cellular immune function against CMV. Therapy primarily involves ganciclovir, now rendered more versatile by data suggesting oral therapy with valganciclovir is not inferior to intravenous therapy with ganciclovir. Treatment of resistant virus remains problematic, but is enhanced by the availability of multiple novel therapeutic agents.

Key words: Cytomegalovirus, diagnostics, infection, prevention, transplant

Abbreviations: CMV, cytomegalovirus; PTLD, post-transplant lymphoproliferative disorder; SOT, solid organ transplant.

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Introduction

Infections remain among the major complications after solid organ transplant, and CMV is the most common of all such infections. Optimal prevention, diagnosis and treatment of active CMV infection are essential, as they significantly impact and enhance transplant outcomes, and will be covered in detail in this section on solid organ transplantation in adults. Recent work allows better understanding of the merits and risks of the main preventative methods, universal prophylaxis and preemptive therapy, allowing programs to choose the best method for their patient

cohorts. Diagnostics have improved significantly, providing more accurate and expeditious methods to diagnose active disease, and also, using novel cellular immune assays, determine the risk of developing either *de novo* or recurrent disease. Intravenous ganciclovir, long the mainstay of therapy, has been partially replaced by oral therapy with valganciclovir, based on recent data demonstrating its therapeutic noninferiority. Treatment of resistant virus remains problematic, but is enhanced by the availability of better diagnostics as well as multiple novel therapeutic agents. Recent guidelines are helpful in guiding optimal management (1–4). These various aspects of CMV management will be covered in this review.

Prevention

Optimal prevention of CMV after transplant can significantly improve overall outcomes. The “direct effects” of CMV (the clinical manifestations) range from asymptomatic viremia to CMV syndrome (fevers, malaise) to invasive disease (colitis, pneumonitis, retinitis, etc). The “indirect effects” of CMV are less clinically prominent, but perhaps more ominous, and augment morbidity and mortality while decreasing graft survival after organ transplant. Patients with active CMV have increased rates of bacterial, viral and fungal infections, posttransplant lymphoproliferative disorder (PTLD), graft dysfunction and failure, acute rejection, chronic allograft nephropathy, interstitial fibrosis and tubular atrophy, transplant and nontransplant vascular disease, diabetes and mortality, as summarized in Table 1 and elsewhere (2,5,6). CMV is immunomodulatory, and can be both immunosuppressive and inflammatory (5). Whether the impact of “indirect effects” correlates with the extent of viremia, or if asymptomatic low-level viremia (as seen with pre-emptive therapy) is sufficient to trigger the “indirect effects” remains to be determined. Optimal prevention of CMV after solid organ transplant (SOT) is essential for all transplant programs that wish to enhance recipient and transplant outcomes.

Universal prophylaxis and preemptive therapy

There are two main methods for CMV prevention: universal prophylaxis and preemptive therapy. Universal prophylaxis involves giving antiviral medication at prophylaxis dose for a defined time to a cohort (i.e. when either donor and/or recipient are seropositive for CMV) or defined subset of a cohort (i.e. given only to the highest risk subset, when donors are seropositive and recipients are negative for CMV,

Table 1: Possible indirect effects of CMV. The relationship between CMV disease and these indirect effects has not been shown in all studies. Citations are examples supporting these statements and are not meant to include all references on this topic. Additional references can be found in the comprehensive review by Freeman (5)

General indirect effects—elevated risks	
Bacterial infections (19,134,135)	
Fungal infection (19,26)	
Viral infections (summarized in (6))	
PTLD (136)	
Cardiovascular events (137)	
New-onset diabetes mellitus after transplantation (138,139)	
Immunosenescence (140)	
Acute rejection (36,37,134)	
Mortality (19,134,141–144)	
Transplant-specific indirect effects	
Chronic allograft nephropathy and/or allograft loss after renal transplant (19,145,146)	
Accelerated hepatitis C recurrence after liver transplant (147)	
Hepatic artery thrombosis after liver transplant (144,148,149)	
Allograft vasculopathy after cardiac transplant (150,151)	
Bronchiolitis obliterans after lung transplant (37,141,143)	

hereafter referred to as D+/R–). Preemptive therapy is defined as serial testing done weekly or biweekly for the first few months after transplant or after treatment of rejection, with treatment dose antiviral therapy initiated once a certain defined positive threshold is reached. While some programs elect not to utilize either of these methods of prevention and to observe clinically with a plan to test and treat based on signs and symptoms of active CMV, this should be generally discouraged, as this is likely to result in high rates of symptomatic CMV disease, and clinical outcomes are likely to be inferior to programs using either preemptive therapy or universal prophylaxis, with more opportunistic infections, higher rates of graft dysfunction and loss and a greater risk of PTLD (6). A retrospective analysis of CMV disease in D+/R+ South African renal transplant patients demonstrated that without any prevention, the incidence of CMV disease was 32%; when valganciclovir prophylaxis was used, the incidence of CMV disease was 4.5% (n = 2/44) (7).

Each method of prevention has merits and disadvantages, as shown in Table 2. While large, randomized studies have not yet been conducted, numerous studies suggest that universal prophylaxis may convey better outcomes compared with preemptive therapy, especially in the higher risk D+/R– population, as summarized in recent guidelines (2); Figure 1 demonstrates the high risk of viremia seen with preemptive therapy in D+/R– recipients in a recent trial, where at 90 days after transplant, 91.9% of D+/R–, 58.9% of D+/R+ and 46.6% of D–/R+ patients had developed viremia. Benefits of universal prophylaxis include fewer opportunistic infections (including Kaposi’s sarcoma and PTLD), improved graft and patient survival,

Table 2: Comparison of known benefits and limitations of prophylaxis versus preemptive therapy (2)

Effect	Prophylaxis	Preemptive
CMV disease	+++	+++
Late CMV disease	++	–
CMV Relapse/treatment failure	++	++
Fewer opportunistic infections	+++	+
Improved graft survival	++	–
Prevention of rejection	++	–
Survival	++	–
Prevention other viruses	+	–
Post transplant lymphoma	+	–
Kaposi sarcoma	+	–
Safety	++	+++
Easier logistics	+++	+
Lower drug cost	+	+++
Lower monitoring cost	+++	+
Resistant CMV	++	++

+ is representative of the ease of use and strength of the evidence, +++ is the strongest evidence or favors the approach listed, a—means no evidence exists. Modified from Kotton et al. (2).

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lower rates of rejection, easier logistics and lower monitoring costs (Table 2). Negatives include higher rates of late CMV, resistant virus and higher drug costs and toxicities. Advantages of preemptive therapy include lower drug cost, reduced drug exposure, lower rates of late CMV (possibly due to enhanced immunologic priming (8)) and theoretically lower risk of resistant CMV due to lower rates of drug exposure. Disadvantages include lower rates of graft and patient survival, higher rates of opportunistic infections, more complex logistics (organizing and managing ~weekly testing for several months after transplant).

Outcomes with preemptive therapy may not be as optimal as with universal prophylaxis, especially in high-risk transplants (i.e. CMV D+/R–), although this is an area of significant controversy, especially in the lower risk seropositive recipients. Controlled trials comparing the two approaches are few. Perhaps some of the more compelling data comes from a randomized clinical trial of oral ganciclovir prophylaxis (N = 74) versus preemptive therapy with intravenous ganciclovir (N = 74), where prophylaxis significantly increased long-term graft survival 4 years after transplant (92.2% vs. 78.3%; p = 0.0425) (9). Those with CMV donor seropositive/recipient seropositive (D+/R+) status had the most dramatic change in rate of graft loss comparing prophylaxis with preemptive therapy (0.0% vs. 26.8%; p = 0.0035). Recent data in 689 renal and liver transplant patients monitored by preemptive therapy demonstrated the effectiveness of this technique in preventing end-organ disease, which occurred in ~1%; viremia occurred in 88% of the D+/R– group, as shown in Figure 1. (10)

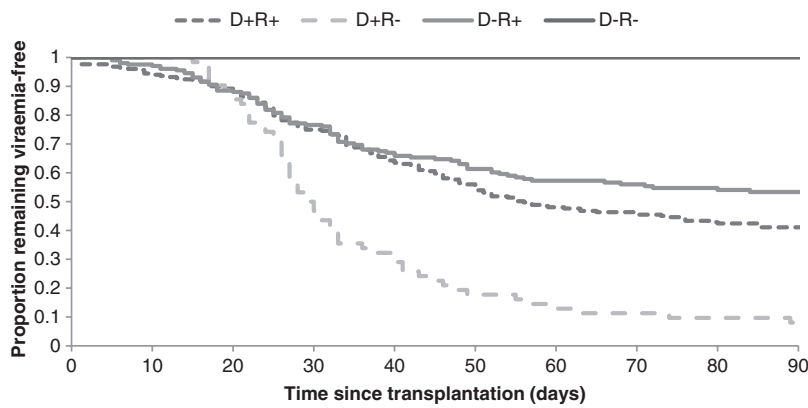


Figure 1: Cytomegalovirus replication kinetics in solid organ transplant recipients managed by preemptive therapy. Kaplan–Meier survival analysis for CMV viremia, indicating the proportion of patients in each of the four DR groups remaining viremia-free through the 90-day follow-up period. $p < 0.0001$ (log-rank test) for a comparison between the groups (10).

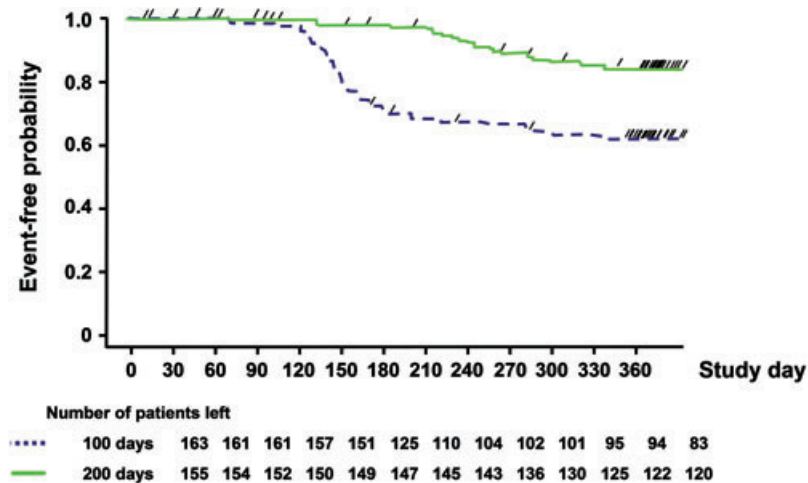


Figure 2: Time to CMV disease in those on 100 days versus 200 days of valganciclovir prophylaxis in the IMPACT study. Kaplan–Meier plot of time to cytomegalovirus disease up to month 12 posttransplant. From (11).

Duration of universal prophylaxis is an important consideration, as longer courses of prophylaxis have been associated with lower rates of late CMV across different types of organ transplants. Some of the more convincing data are in the IMPACT trial (11), in high-risk (i.e. CMV D+/R–) kidney transplant recipients enrolled in an international, multicenter, double-blind, randomized controlled trial comparing the efficacy and safety of 200 days versus 100 days of valganciclovir prophylaxis in 326 high-risk (CMV D+/R–) kidney allograft recipients. Significantly fewer patients in the 200-day group versus the 100-day group developed confirmed late CMV disease by 12 months after transplant (16.1% vs. 36.8%; $p < 0.0001$), as seen in Figure 2, which was later confirmed in 2 years of follow-up (21.3% vs. 38.7%; $p < 0.001$) (12), and CMV viremia was also significantly lower in the 200-day group (37.4% vs. 50.9%; $p = 0.015$ at month 12). There was no significant difference in the rate of biopsy-proven acute rejection between the groups (11% in 200 day cohort vs. 17% in 100 day cohort, $p = 0.114$). There was a much lower risk of opportunistic infections in those given 200 days of prophylaxis (13% vs. 27%, $p = 0.001$). Thus, extending valganciclovir prophylaxis to 200 days significantly reduced

the incidence of CMV disease and viremia compared with 100 days” prophylaxis, without significant safety concerns associated with longer treatment. Similarly, in a multicenter, placebo-controlled trial involving 11 United States lung transplant centers, Palmer et al. (13) evaluated 66 lung transplant recipients who completed 3 months of valganciclovir prophylaxis and compared them with 70 who were given 12 months of prophylaxis. CMV disease occurred in 32% of the shorter course group versus 4% of the extended-course group ($p < 0.001$). Significant reductions were observed with respect to severity of CMV infection, peak viremia and disease severity with extended prophylaxis. Rates of acute rejection, opportunistic infections, CMV UL97 ganciclovir-resistance mutations, adverse events and laboratory abnormalities were similar between the two groups. A single-center observational study of 128 lung transplant recipients on indefinite valganciclovir prophylaxis, regardless of donor or recipient CMV serostatus, found no significant difference in rates of CMV disease between those who were on continuous prophylaxis or not (4.6% vs. 4.9%; $p = 1$), but an increased incidence of laboratory-detected CMV infection in those who discontinued prophylaxis (40.7%

vs. 12.7%; $p = 0.001$) (14). Sixty-five patients (50.6%) discontinued valganciclovir prophylaxis, either temporarily or permanently, primarily due to mild leukopenia.

Costs have a significant influence on transplant care worldwide, and may significantly influence the choice of prevention method. Costs of serial testing for preemptive therapy (including personnel and laboratory monitoring costs) and of medications for universal prophylaxis vary across different settings, as do the net costs to the patient (i.e. copayments for medication) or to the transplant program or healthcare system. In one recent modeling study, the incidence of CMV infection in seropositive kidney transplant recipients was 4.1% and 55.5% within the first year after transplant while under universal prophylaxis and preemptive therapy, respectively (15). Universal prophylaxis incurred \$1464 more in direct cost compared with preemptive therapy, while saving \$7309 in indirect cost, and resulted in a net gain of 0.209 in quality-adjusted life years per patient over a 10-year period. Thus, universal prophylaxis resulted in a cost saving of \$27 967 per quality-adjusted life year gained when compared with preemptive therapy. Using data from the Improved Protection Against CMV in Transplant (IMPACT) trial demonstrated that prolonged prophylaxis of 200 days with valganciclovir compared with 100 days significantly reduces the incidence of CMV in high-risk kidney transplant (D+/R-) recipients; a cost-effectiveness model was developed to evaluate prolonged prophylaxis (200 days) with valganciclovir and its long-term economic impact from the US healthcare payer perspective (16). For the first 5 years, they found that the incremental cost-effectiveness ratio of US \$14 859/quality-adjusted life year suggests that 200-day valganciclovir prophylaxis is cost effective compared with the 100-day regimen, considering a threshold of US \$50 000 per quality-adjusted life year. The 10-year analysis revealed the 200-day prophylaxis as cost saving with a 2380 quality-adjusted life year gain (per 10 000 patients) and simultaneously lower cost. They concluded that prolonged prophylaxis with valganciclovir reduces the incidence of events associated with CMV infection in high-risk D+/R- kidney transplant recipients and is a cost-effective strategy in CMV disease prevention. Another single-center, retrospective study similarly concluded that 6 months of prophylaxis in those who are CMV D+/R- was more cost effective than 3 months, with an incremental cost of \$34 362 and \$16, 15 per case of infection and disease avoided, respectively, and \$8304 per one quality adjusted life-year gained (17). Thus, data suggest that universal and longer prophylaxis is more cost effective with respect to long-term outcomes.

Late CMV remains a significant problem in those recipients on universal prophylaxis, which sometimes delays but does not prevent CMV infection (either primary infection in seronegative recipients, or less commonly, reactivation disease in seropositive recipients) once the antiviral pro-

phylaxis is stopped. In one series, 47/127 (37%) D+/R- kidney transplant recipients developed late CMV disease after the prophylaxis was stopped, at a median 244 days after transplantation (range 150–655) and median 67 days after the cessation of prophylaxis (range 1–475); this high rate of late CMV disease lead the authors to conclude that CMV primary infections were common after 6 months of valganciclovir prophylaxis and mostly symptomatic, with relapses common, suggesting that the prevention strategies for CMV need to be reconsidered (18). In another series of D+/R- kidney recipients, 51 patients (29%) developed CMV disease at a median of 61 days (interquartile range, 40–143 days) after stopping 3 months of antiviral prophylaxis (19). In a third series of D+/R- renal transplant recipients on 3 to 6 months of antiviral prophylaxis, CMV disease developed in 29 of 113 (26%) at a median of 185 days posttransplant (range 116–231 days), including CMV syndrome (66%) and tissue invasive disease (34%) (20). All series have a relatively high rate of late CMV, underscoring the ability of preventative antiviral therapy to delay but not prevent CMV infection. Whether longer therapy (> 6 months) after renal transplant might better prevent late CMV remains to be studied; some experts recommend longer or indefinite prophylaxis in higher risk patients (21,22).

Recurrent disease after preemptive therapy or treatment of primary infection is not rare, especially in D+/R- SOT recipients. Whether to initiate secondary chemoprophylaxis or viral monitoring after treatment of CMV infection has not been well studied. In a study of D+/R- subjects who received 3–6 months of antiviral prophylaxis followed by weekly viral loads for 8 weeks (considered a ‘hybrid approach’, combining universal prophylaxis with subsequent preemptive therapy), symptomatic CMV disease occurred in 29 of 71 (40.8%) patients during the first year; viremia occurred in 19 of 71 (26.8%) patients during the 8-week surveillance, but a significant portion ($n = 16$) occurred after the 8-week surveillance period, suggesting that monitoring in high-risk recipients after prophylaxis was of limited value due to rapid viral doubling time (median doubling time 1.1 days) and disease occurring after the surveillance period (23). Experts vary in their practices, and recommend that institutions develop local protocols, based on previous clinical outcomes, use of cytolytic induction therapies, the net state of immunosuppression, type of organ transplanted, costs, testing ability and other factors (2).

The choice of immunosuppressive regimen may alter the risk of CMV disease. Sirolimus has antiviral properties and conveys a lower risk of CMV disease (24–27); one prospective study of 1470 renal transplant recipients (55 of whom were kidney–pancreas transplant recipients) found that the use of sirolimus had a protective effect against CMV disease (odds ratio 0.27) (24), the Symphony trial also found a much lower rate of CMV in those on low dose sirolimus (6%) compared with those on standard or low cyclosporine

A (15% and 11%) ($p = 0.003$) (28), and another trial noted that everolimus conveyed a reduction in the incidence of CMV infection, syndrome and viremia in *de novo* renal transplant recipients (29).

Antiviral medications for universal prophylaxis and preemptive therapy

There are three main agents commonly used for universal prophylaxis and preemptive therapy: intravenous ganciclovir, oral valganciclovir and oral ganciclovir. In general, the dose for universal prophylaxis is half that of preemptive therapy (which is standard treatment dose). Foscarnet and cidofovir are very rarely used for routine prophylaxis, as they have significant toxicities and require close clinical monitoring; additionally, they are only available in an intravenous formulation. Acyclovir and valacyclovir are also used to prevent CMV, although both have significantly reduced anti-CMV activity and are not recommended as first-line agents in recent guidelines (1–4), but are effectively used to prevent other human herpesvirus infections, including varicella, and as such have an important role in antiviral prophylaxis for CMV D–/R– SOT recipients. In early work on CMV prevention, acyclovir was determined to be inferior to oral ganciclovir (30), although at lower doses than others have used. Meta-analysis of direct comparison studies demonstrated that ganciclovir was more effective than acyclovir in preventing CMV disease (7 studies; RR 0.37, 95% CI 0.23–0.60) (31). In initial studies, valacyclovir doses were quite high (8 g/day) although more recent work favors lower doses (3 g/day, renally adjusted) (32); similarly, the dose of acyclovir is also high (3.2 g/day). Clinicians have been concerned about nephrotoxicity and neurotoxicity (30) with such high doses, and most prefer to reserve acyclovir and valacyclovir for herpes simplex and zoster prevention in CMV D–/R– recipients (2).

Valganciclovir has revolutionized both preventative and therapeutic options, as it is approximately 60% bioavailable (33), almost 10-fold more than oral ganciclovir (34). In the PV16000 study, Paya et al. (35) compared the efficacy and safety of valganciclovir versus oral ganciclovir for preventing CMV disease in high-risk D+/R– SOT recipients. In this randomized, prospective, double blind, double-dummy study, 364 CMV D+/R– patients received valganciclovir 900 mg once daily or oral ganciclovir 1000 mg three times a day starting within 10 days of transplant and continued through 100 days. By 12 months, CMV disease developed in 17.2% and 18.4% of valganciclovir and ganciclovir patients, respectively, and the incidence of investigator-treated CMV disease events was comparable in the valganciclovir (30.5%) and ganciclovir (28.0%) arms. CMV viremia during prophylaxis was significantly lower with valganciclovir (2.9% vs. 10.4%; $p = 0.001$), but was comparable by 12 months (48.5% valganciclovir vs. 48.8% ganciclovir). Valganciclovir delayed the onset of CMV disease and viremia. Rates of acute allograft rejection were lower with valganciclovir. The safety profile was similar for

both drugs, except for a higher incidence of neutropenia with valganciclovir (8.2%) versus ganciclovir (3.2%). Overall, PV16000 showed that once daily oral valganciclovir was noninferior and as clinically effective and well tolerated as oral ganciclovir for CMV prevention in high-risk SOT recipients. A trial of valganciclovir versus oral ganciclovir in lung transplant recipients showed a higher rate of bronchiolitis obliterans syndrome and bacterial tracheobronchitis in those on oral ganciclovir (36); another similar trial showed much lower rates of CMV pneumonitis in those on valganciclovir compared with oral ganciclovir prophylaxis (37).

Antiviral therapy for CMV should always be carefully adjusted to renal function. Use of low-dose antivirals increases the risk of treatment failure and development of antiviral resistance, and should be avoided (2). Treating clinicians should calculate the renal function and refer to the package insert (see Table 3) for optimal management. For kidney recipients with a creatinine clearance of 40–59 mL/min, the correct dose of valganciclovir for prophylaxis would be 450 mg a day. Higher doses are unlikely to convey benefit, and may result in a greater likelihood of neutropenia and side effects, in addition to higher costs.

Optimal antiviral(s) for universal prophylaxis may vary by type of organ transplanted. Some experts tend to use intravenous ganciclovir (rather than valganciclovir) for the more immunocompromised transplant recipients, such as lung or heart transplant recipients (2). Valganciclovir was not approved in the United States for prophylaxis in liver transplant recipients, based on a small subgroup analysis of the PV16000 data (35), which looked at tissue-invasive disease and then subsequently analyzed the small liver subpopulation of this subgroup; there are statistical shortcomings to this approach. Valganciclovir is approved for prophylaxis in liver transplant patients in the European Union and Canada. The majority of clinicians polled use it for prophylaxis (38). There are limited data as far as the optimal prophylactic agent after liver transplant. In one study of 66 D+/R– liver transplant recipients at a single institution who were given one of three prophylactic regimens: valganciclovir (900 mg daily; 27 patients), oral ganciclovir (1000 mg every 8 h; 17 patients) or intravenous ganciclovir (6 mg/kg daily; 22 patients) (39), eight CMV cases occurred, all after completion of the prophylaxis. CMV disease occurred in 22% of valganciclovir recipients, 5% of intravenous ganciclovir recipients and 6% of oral ganciclovir recipients, leading the authors to conclude that the fourfold higher incidence of CMV disease supports the avoidance of valganciclovir for prophylaxis in high-risk OLT patients.

CMV immunoglobulin was more frequently used for prophylaxis prior to the advent of valganciclovir. A meta-analysis of 11 randomized trials (698 patients; median follow-up: 12 months, range: 3–22 months), with six randomized trials (302 patients) after kidney transplantation, demonstrated a beneficial effect of the prophylactic use of CMV immunoglobulin on total survival and prevention of

Table 3: Optimal dosing of ganciclovir and valganciclovir, with adjustments for renal function. Taken from Kotton 2010 (152), original references (33,34,153)

CrCl (mL/min)	Induction dose	Maintenance/prevention dose	Comments
Intravenous ganciclovir (153) from http://www.gene.com/gene/products/information/cytovene/pdf/pi.pdf			
≥70	5.0 mg/kg IV q12 h	5.0 mg/kg IV q24 h	Requires IV access; leukopenia common
50–69	2.5 mg/kg IV q12 h	2.5 mg/kg IV q24 h	
25–49	2.5 mg/kg IV q24 h	1.25 mg/kg IV q24 h	
10–24	1.25 mg/kg IV q24 h	0.625 mg/kg IV q24 h	
<10	1.25 mg/kg IV three times a week after hemodialysis	0.625 mg/kg IV three times a week after hemodialysis	
Valganciclovir (33) (from http://www.gene.com/gene/products/information/valcyte/pdf/pi.pdf)			
≥60	900 mg twice daily	900 mg once daily	Easy to administer; leukopenia common
40–59	450 mg twice daily	450 mg once daily	
25–39	450 mg once daily	450 mg every 2 days	
10–24	450 mg every 2 days	450 mg twice weekly	
<10	Not recommended	not recommended	
Oral ganciclovir (34) from http://www.drugs.com/mmx/cytovene.html			
≥70	Should not be used	1000 mg three times a day with food, or 500 mg six times a day every 3 h with food, during waking hours	Low oral bioavailability; high pill burden
50–69	Should not be used	1500 mg once a day, or 500 mg three times a day	
25–49	Should not be used	1000 mg once a day, or 500 mg twice a day	
10–24	Should not be used	500 mg once a day	
<10	Should not be used	500 mg three times a week, following hemodialysis	

CMV-associated death in SOT recipients (although not kidney transplant recipients). CMV disease was significantly reduced in all recipients receiving prophylactic CMV immunoglobulin, although it had no impact on CMV infections and clinically relevant rejections (40). A recent analysis of Scientific Registry of Transplant Recipients (SRTR) data in the United States suggests that in pediatric heart transplant recipients, CMV immunoglobulin (with or without antivirals) and antivirals without CMV immunoglobulin were both associated with significantly ($p \leq 0.05$) lower rates of graft loss and death versus no prophylaxis (41). Although there are limited data to support its use in the era of more potent antiviral prophylaxis, current recommendations tend to include CMV immunoglobulin use in higher risk organ transplant recipients, including lung, heart and intestinal transplant recipients (2), especially in those who are D+/R–.

Leukopenia is common in transplant recipients due to multiple bone marrow suppressive medications, including cytolytic induction therapies, mycophenolate mofetil, (val)ganciclovir and others. In the IMPACT study, the overall reported incidence of leukopenia was 38% in the 200-day group versus 26% in the 100-day group; however, the median laboratory white blood cell counts and the incidences of reported neutropenia, febrile neutropenia, agranulocytosis, anemia, thrombocytopenia and pancytopenia were comparable between the two groups (11). Guidelines suggest that management of leukopenia include reduction of other bone marrow suppressive medications, and either stopping or continuing but not dose reducing (val)ganciclovir (2,4). When the antiviral prophylaxis is stopped, clinicians may wish to switch to preemptive therapy.

Vaccines and tailored prevention based on immunology: the way of the future?

Vaccination against CMV has been an elusive goal, both for developing a primary immune response in those naïve to CMV, or augmenting immunity in seropositive recipients. An effective vaccine could revolutionize the impact and management of CMV on SOT. Additional details on CMV vaccines against CMV are covered in the section by V. Emery, pp. 79.

Development of an immune response to CMV in naïve SOT recipients is crucial toward further control of the virus. Preemptive therapy allows for viral replication and priming of the immune system, and there may be a therapeutic effect of the acquisition of CMV-specific immune response. In a study of renal transplant recipients analyzed for CMV viremia and CMV-specific T-cell response (by interferon-gamma enzyme-linked immunospot assay) before transplantation and at 30, 60, 90, 180 and 360 days after transplantation, CMV-seropositive transplant recipients displayed progressive immune reconstitution starting from day 60 after transplantation (8). CMV-seronegative recipients did not mount a detectable T-cell response throughout the time on prophylaxis, whereas a single episode of CMV viremia in those on preemptive therapy was sufficient to prime a protective T-cell immune response. The authors concluded that baseline immunity and antiviral therapy but not antithymocyte globulin treatments profoundly influence T-cell reconstitution in kidney transplant recipients. Another study of 21 subjects on preemptive therapy after SOT, who developed CMV replication episodes at a median of 4 weeks (range 2–8 weeks) after transplantation and a CMV-specific T-cell response (as determined by CMV-specific T-cell intracellular cytokine

staining) at a median of 12 weeks (range 10–20 weeks), experienced a decline in the incidence of CMV replication episodes that was inversely correlated with the acquisition of the CMV-specific T-cell response (42). In addition, after acquisition of the immune response, 42 CMV replication episodes were cleared without treatment. The authors did not comment on long-term graft outcomes or on the indirect effects of CMV. Other groups report that both immune response and viremia should be monitored while on pre-emptive therapy to determine risk of infection (43,44). Both CMV-specific CD4+ and CD8+ T cells are needed for protection against CMV disease (45).

Optimally, the length of prophylaxis would be tailored to the individual patient. CMV-specific cellular immune responses may correlate with the risk of CMV disease posttransplant, thus may be clinically useful in guiding prevention (43). Such assays include tetramer staining and intracellular cytokine staining for IFN- γ (both using flow cytometry), bead-based cytokine profiling, ELISpot, QuantiFERON[®]-CMV (Cellestis Ltd., Melbourne, Australia) and ATP-release assay (i.e. Immuknow[®], (Cylex, Colombia, MD, USA) which is not specific for CMV). Most studies that have analyzed CMV-specific T-cell responses have used intracellular cytokine staining for IFN- γ using flow cytometry (available primarily in research settings) to demonstrate that low levels of CMV-specific cellular immunity predict an increased risk of CMV disease (46–48) and inability to clear viremia (46,49,50). The predictive value for viremia may be improved when the analysis of IFN- γ is combined with other cytokines such as interleukin-2 and additional markers such as programmed death-1 mutations to predict viremia and CMV disease (51,52). For additional discussion on immunologic aspects of CMV, please refer to section on “Immune Regulation of Human Herpesviruses and Its Implications for Human Transplantation” by Corey Smith and Rajiv Khanna, pp. 9.

A limited number of such assays are commercially available. QuantiFERON[®]-CMV, an ELISA-based assay, is approved in the European Union; the Immuknow[®] assay is FDA approved. Kumar et al. used the QuantiFERON[®]-CMV, which reflects cellular immunity to CMV via quantification of the interferon-gamma response after whole-blood stimulation with a 21-peptide pool, to show that monthly evaluation of 108 SOT recipients of mixed serostatus detected cell-mediated immunity in 38/108 (35.2%) patients (48). Those with a detectable interferon-gamma response had lower rates of CMV disease (2/38 (5.3%) patients) compared with those recipients with a negative interferon-gamma response, who had disease in 16/70 (22.9%) ($p = 0.038$), suggesting that such monitoring may be a useful tool for predicting late-onset CMV disease. Work in allogeneic stem cell transplant recipients suggests that the QuantiFERON[®]-CMV is somewhat less sensitive than intracellular cytokine staining (53). At present, there are no randomized, multicenter data suggesting that clinical management decisions based on immunologic monitoring af-

fect patient outcomes, although this is a rapidly evolving area of research.

Diagnosis

Diagnostic tests for CMV include serology, reflecting prior exposure; tests for active disease, including quantitative nucleic acid testing (QNAT), antigenemia, culture and histopathology, as well as newer immunology assays, reflecting the cellular immune response to CMV. Prior to transplant, CMV serology should be performed on all donors and recipients. Tests for anti-CMV IgG are recommended, as they have better specificity than IgM or combination IgG and IgM tests, neither of which should be used for pretransplant screening, as false positive tests for IgM may significantly decrease test specificity (54–56). If the initial result was negative and there was a significant lapse in time after testing, serology should be repeated at the time of SOT, in order to not overlook those donors or recipients who are now seropositive. Confounders of CMV serology testing include low immunoglobulin states and plasmapheresis (more likely to be false negative) or by transfusion of blood products (more likely to be false positive via passive transfer of antibody), thus a pretransfusion test result (or sample) may be more accurate. Equivocal or borderline serology results occur infrequently. The Transplantation Society guidelines recommend for adults that an equivocal serology result should be assumed to be positive in a donor but negative in a recipient, an approach which ensures that the recipient is assigned to the highest appropriate CMV risk group for posttransplant management decisions (2). Neither IgG nor IgM serology has ever been shown to accurately reflect active disease, and are not considered helpful in the diagnosis of active CMV disease after SOT.

Late seroconversion (i.e. after the end of prophylaxis) may be useful for identifying those patients at lower risk of late-onset CMV disease; in a trial of 352 D+/R– transplant recipients that compared 100 days of ganciclovir with valganciclovir prophylaxis, while seroconversion at the end of prophylaxis (day 100) was not predictive of subsequent CMV disease (CMV disease 13.3% if seropositive vs. 17.8% if seronegative; $p = \text{NS}$), negative IgG serostatus 6 months posttransplant was predictive of subsequent CMV disease in the ensuing 6 months (CMV disease 1.3% if seropositive vs. 10.0% if seronegative; $p = 0.002$) (57). In the IMPACT trial, which compared 318 CMV D+/R– kidney transplant recipients receiving valganciclovir once daily for 100 days versus 200 days, IgM or IgG seroconversion was delayed and somewhat attenuated in the 200 day group at 55.5% vs. 62.0% in the 100-day group by 2 years; $p = 0.26$ (12). Seroconversion was very rare while on prophylaxis, but all patients who had CMV disease seroconverted; 7.7% (7 of 91) at the onset of disease, and 92.3% (84 of 91) after disease onset. Interestingly, a number of

patients seroconverted without CMV disease (24.5% in the 100-day group vs. 32.9% in the 200-day group).

Viremia is most commonly detected by either an antigenemia assay or a QNAT test. The original test for viremia, CMV pp65 antigenemia, is a semiquantitative test that has been shown to be helpful in initiating preemptive therapy, the diagnosis of clinical disease, and monitoring response to therapy (58–62). It is relatively easy to perform and does not require expensive equipment, although there are problems with a lack of assay standardization, including subjective result interpretation. Limitations include neutropenia (the assay cannot be performed with absolute neutrophil counts less than 1000 neutrophils/uL), and limited stability of the blood specimen, which should be processed within 6–8 h of collection.

CMV QNAT is the main alternative option to antigenemia (63–69). The testing requires expensive equipment and specialized expertise. Since the viral load can vary significantly between plasma and whole blood specimens (CMV DNA is detected earlier and usually in greater quantitative amounts in whole blood), one specimen type should be used when serially monitoring patients (70–73). QNAT results can vary widely across different testing centers, due to variation in nucleic acid extraction, assay design and until recently, the lack of an international reference standard (74), such that results from different testing sites cannot be compared (i.e. to determine whether viral load rising or falling), and has prevented the establishment of broadly applicable thresholds for clinical decision making. Transplant centers should use one testing modality and compare results within that one rubric (2).

A multicenter, international study assessing the variability of CMV viral load testing across 33 laboratories demonstrated that the inconsistency in viral load values for individual samples ranged from 2.0 to 4.3 log₁₀ copies/mL, such that a CMV QNAT of 100 copies/mL in a laboratory may be reported as 100 000 copies/mL (3 log₁₀ difference) in another laboratory. Variation was greatest at lower values, and less with commercially available reagents and procedures (74).

Fortunately, an international standard for CMV was developed and approved in November 2010 by the World Health Organization (75). This international standard, composed of a standardized quantity of CMV, will allow laboratories and manufacturers to assess the accuracy of viral load values and to calibrate different individual assays. Although still in the early stages, this standard will lead to harmonization of viral load values between laboratories, such that results can be reported as international units (IU) (rather than copies) per milliliter and compared across various testing platforms (76).

Factors influencing choice of test include available resources, technical expertise, patient population (and how

far away they live), turnaround time, volume of samples tested and cost. Neither test has been shown to be clinically superior. Both the antigenemia and QNAT viral load tests have been shown to have excellent clinical utility; correlation between QNAT and CMV antigenemia levels is not always consistent (63,77,78).

One specimen type (i.e. plasma versus whole blood) should be used for serial viral load testing, since the results vary across different specimen types (2). Recent work in patients on treatment of CMV disease compared viral load testing of plasma versus whole blood real-time PCR demonstrated good correlation but significant differences in absolute value and clearance kinetics (79). Virus was still detectable by day 21 in 154 of 219 (70.3%) patients by whole blood test, versus 105 of 219 (52.1%; $p < 0.001$) patients with the plasma assay, with similar positive predictive values for virologic recurrence. In the subset of patients with negative plasma but positive whole blood at day 21, the incidence of virologic recurrence was the same as that of all patients with a negative plasma assay (23% in each group). The authors concluded that when treating CMV disease, enhanced detection of residual viremia using a whole blood real-time PCR does not seem to offer significant clinical advantages nor allow for better prediction of recurrence of CMV viremia or disease, and suggest that the treat-to-negative paradigm may not hold true when using such highly sensitive whole-blood assays.

Other body fluids and tissues, including biopsy, bronchoalveolar lavage and CSF specimens, can be tested for CMV by QNAT, which may improve sensitivity, potentially with faster results compared to culture (14,80,81). Several studies suggest QNAT on bronchoalveolar lavage specimens may be helpful in predicting pneumonitis (81–83), although not in all studies (14). The presence of CMV DNA in the CSF likely represents CMV disease and should be treated. Diagnostically, retinitis is based on clinical ophthalmologic examination; assays for viremia in blood are rarely useful as predictors of CMV eye disease; this absence of viremia may also be seen with colitis and esophagitis.

CMV culture can be slow, expensive and less sensitive. Seropositive humans may shed CMV in their secretions, especially during times of stress, rendering positive cultures that do not necessarily reflect active disease. Viral culture of blood for CMV has poor sensitivity, while CMV urine, stool and sputum cultures have poor specificity (84). Culture of tissue specimens remains an important option for diagnosis of tissue invasive disease, particularly for gastrointestinal samples (i.e. colonic biopsies), where antigenemia or polymerase chain reaction (PCR) testing on blood may not always be positive even with invasive disease.

Immunohistochemistry for CMV should be routinely performed on all biopsy specimens where CMV is suspected, to maximize diagnostic sensitivity. Identification of inclusion bodies or viral antigens in biopsy material (82,85) or

in bronchoalveolar lavage specimens cells is very specific for CMV disease, especially with a positive culture. Tissue invasive CMV disease, such as colitis or hepatitis, should be confirmed by immunohistochemistry or *in situ* DNA hybridization (86–88). Different antibodies have variable sensitivity, and results may vary between fresh and formalin fixed paraffin-embedded tissue (88).

Therapy

Treatment of CMV infection may include antiviral therapy (sometimes followed by prophylaxis and/or monitoring), reduction of immunosuppression and occasional use of CMV immunoglobulin. Valganciclovir and intravenous ganciclovir are the first line agents used to treat CMV disease. Increasingly, valganciclovir has become the preferred agent used to treat SOT recipients with mild-to-moderate CMV infection; the VICTOR trial demonstrated that oral valganciclovir is noninferior to intravenous ganciclovir for treatment of CMV disease in SOT recipients (74% renal transplant recipients) with generally nonlife-threatening disease (48% had CMV syndrome and 49% had tissue-invasive CMV disease) (7). A total of 321 SOT recipients with non-life-threatening CMV disease were treated with either oral valganciclovir 900 mg twice daily or intravenous ganciclovir 5 mg/kg twice daily (both renally adjusted) for 21 days, followed by preventative valganciclovir 900 mg daily in both arms for 28 days. CMV viral load declined at the same rate in both groups. Viremia eradication at day 21 was 45.1% for valganciclovir and 48.4% for ganciclovir, and by day 49 67.1% and 70.1%, respectively, neither of which were statistically different. Treatment success, as assessed by investigators, was similar in both groups, 77.4% versus 80.3% at day 21 and 85.4% versus 84.1% at day 49. Side effects and treatment discontinuations were comparable. Oral valganciclovir was thus shown to have comparable safety and noninferiority to intravenous ganciclovir for treatment of CMV disease in a subset of SOT recipients with mild-to-moderate disease. In patients with severe or life-threatening CMV disease, intravenous ganciclovir is still the preferred drug, as oral treatment has not been documented to provide equivalent treatment; it should be used in patients that do not tolerate oral treatment or have suboptimal absorption of valganciclovir. Oral ganciclovir is not recommended or approved for treatment due to low bioavailability. Correct dosing of valganciclovir/ganciclovir to renal function limits toxicity (89) (see Table 3 for dosing); subtherapeutic dosing increases the risk of clinical failure and antiviral resistance (90).

Viremia should be tested weekly on therapy. Recent guidelines and data have suggested that treatment be continued until two weekly assays of active viral replication (i.e. CMV viral load or CMV pp65 antigenemia) are negative, reflecting the “treat-to-negative paradigm”, or for a minimum of 2 weeks; therapy can then be stopped com-

pletely, or switched to prophylaxis dosing (discussed below) (2,3,91–93). Duration of treatment based on response to treatment is likely to be superior, compared with a standard duration of treatment. Data from the VICTOR trial suggests that stopping therapy at day 21 in those who did not have eradication of CMV viremia resulted in high rates of disease recurrence (93). In addition, it demonstrated that high initial viremias were less likely to have cleared after 21 days of treatment, suggesting that treatment of patients with high viral loads should generally be treated longer than those with a lesser initial burden of disease. Data on the clinical utility of whole blood versus plasma CMV viral load assays for monitoring the therapeutic response suggests that one specimen type be used for serial testing, due to variation among sample types; neither was clinically superior (79).

Treatment of intestinal disease, the most common manifestation of invasive CMV (18), can be more complicated, as viremia may be low or even negative in the setting of significant gut disease. A recent study of clinical predictors of CMV relapse after treatment of primary gastrointestinal CMV disease in SOT recipients found that extensive involvement of the gastrointestinal tract was significantly associated with CMV relapse, occurring in 27%, while CMV seroconversion, viral load, treatment duration, maintenance therapy and endoscopic findings at the end of therapy were not significantly associated (94). The median time to CMV PCR negativity in blood was 22.5 days (range, 16.5–30.7) and to normal endoscopic findings was 27.0 days (range, 21.0–33.5), suggesting that for optimal treatment of intestinal disease, additional antiviral treatment after clearance of viremia is needed.

Relapse of CMV disease after treatment is frequent; optimal methods to mitigate risk are not well studied. Secondary prophylaxis with antivirals for 1–3 months after treatment varies considerably across transplant centers, but is frequently employed, especially when the risk of relapse is high (2,7). Close clinical and microbiologic monitoring (i.e. preemptive therapy, as discussed above in the Prevention section) in the first few months after treatment remains another commonly used option. Some groups use a hybrid approach, combining various methods for secondary prevention. Those at higher risk for relapse include those with primary CMV infection (D+/R–), deceased donor transplantation, high baseline viral load, persistent viremia when transferred to secondary prophylaxis, multi-organ disease and treatment of rejection (1,92,93,95,96). In a recent retrospective study of 1760 SOT recipients, 105 cases of CMV viremia occurred, with relapse occurring in 19%, of which 50% had end-organ disease; multivariable analysis identified risks factors for relapse including thoracic organ transplant and diabetes mellitus (97).

Reduction of the immunosuppression should be considered with severe CMV disease, with very high viral loads, in clinically refractory disease and with leukopenia (98);

it may also be advisable in less severe cases of CMV infection. Clinicians may wish to return to standard immunosuppressive treatment when adequate clinical and viral response is obtained. Late or recurrent CMV disease, especially more than 1 year after SOT, may suggest overimmunosuppression. Data on the effects of the intensity of immunosuppressive therapy on the outcome of treatment for CMV disease in organ transplant recipients from the VICTOR trial suggest that better early viral load eradication occurred with dual versus triple immunosuppressive therapy, lower blood concentrations of calcineurin inhibitors, and longer time since transplantation (99). The type of calcineurin inhibitor (tacrolimus/cyclosporine) or use of mycophenolate did not affect treatment efficacy, although both tacrolimus- and mycophenolate-treated patients showed a lower rate of CMV recurrence. Lower total intensity of immunosuppressive therapy was associated with more effective early, but not overall, viral load eradication on valganciclovir/ganciclovir therapy (97).

The role of CMV immunoglobulin in the treatment of CMV disease is unclear; it is sometimes used as adjunctive therapy for severe forms of CMV disease, including pneumonitis or with resistant virus (2). While still experimental, adoptive infusions of CMV-specific T cells may prove helpful in certain situations (100,101).

Antiviral resistance

Antiviral resistance, most commonly to ganciclovir, remains an Achilles heel of CMV treatment and is associated with higher morbidity and mortality. Outcomes range from asymptomatic to severe or fatal disease (102–105). Antiviral resistance is associated with lower doses and/or prolonged use of antiviral agents, D+/R– transplants, higher immunosuppression, severe tissue-invasive CMV disease and/or high viral loads, and lung transplantation (105–108). In a recent study of the incidence and outcomes of ganciclovir-resistant CMV viremia in 1244 kidney recipients transplanted at a single center from 2004 through 2008, 26 of 27 cases were in the D+/R– transplant recipients (107); in a French national cohort of 346 with resistant virus, 42% were D+/R– (108).

Both universal prophylaxis and preemptive therapy have been reported to increase the risk of resistant CMV. In one single-center series of 225 CMV D+/R– transplant patients who received valganciclovir prophylaxis for a median of 92 days, 65 (29%) of the 225 patients developed late primary CMV disease, including nine (14%) suspected to have drug-resistant virus, of whom 4 (6.2%) had confirmed UL97 or UL54 mutations and elevated rates of allograft loss and mortality (109). Similarly, a trial of 128 lung transplant recipients on indefinite valganciclovir prophylaxis had a 2.3% ($n = 3$) rate of CMV resistance, still relatively low but significant (14). In a trial comparing 32 D+/R– kidney transplant recipients who received 3 months of valganciclovir prophylaxis with 80 D+/R– KTR who received pre-

emptive treatment, UL97 or UL54 mutations were more frequent in the preemptive group (16% vs. 3% in the prophylaxis group; $p = 0.05$) (110). In the above-mentioned study of 1244 kidney recipients on preemptive therapy with low dose VGCV (900 mg once daily, renally adjusted), a high incidence of CMV UL97-resistance gene mutations in D+/R– patients was found (107). On the other hand, antiviral resistance was not seen in two recent prophylaxis trials: the IMPACT trial (11), the international, multicenter, double-blind, randomized controlled trial comparing the efficacy and safety of 200 days versus 100 days of valganciclovir prophylaxis in D+/R– kidney transplant recipients or the lung transplant trial by Palmer et al. (13) comparing 3 versus 12 months of valganciclovir prophylaxis which demonstrated no elevated risk of ganciclovir-resistant CMV with prolonged prophylaxis. Similarly, in a French cohort of SOT recipients with resistant CMV, prophylaxis (acyclovir, valacyclovir or valganciclovir) was not significantly associated with virologic resistance when compared with preemptive treatment ($p = 0.55$) (108). Contradictory data make it hard for clinicians to decide which prevention method is preferable with respect to resistant CMV.

The diagnosis of ganciclovir resistance is based on the lack of clinical or virologic response to therapy. On treatment, symptoms and CMV viremia usually recede fairly quickly over 2 to 3 weeks; when a plateau or lack of response is noted, clinicians should consider further evaluation and treatment for resistant virus. The advent of commercially available PCR-based genotyping provides rapid data on the exact UL97 or UL54 mutation(s) seen, and can be very helpful in guiding therapeutic decisions (111). The plaque reduction (phenotypic) sensitivity assay is fairly impractical for routine patient care, because of slow turnaround time, the need to culture virus, technical complexity, and difficulties with standardization.

More than 90% of ganciclovir-resistant CMV isolates contain viral phosphotransferase (UL97) mutations, and the rest are found primarily in the viral DNA polymerase gene UL54 gene. Well-characterized CMV drug resistance mutations has been described (112–115). In patients treated with ganciclovir, UL97 mutations almost always appear first, followed later by the addition of UL54 mutations that confer increased ganciclovir resistance and cross-resistance to cidofovir or foscarnet (108,116–118).

Guidelines for managing resistant CMV suggest either increasing the dose of ganciclovir with mild-to-moderate disease, or switching to foscarnet with more severe and/or visually threatening disease, with or without continued use of ganciclovir (2,119). Cidofovir is not usually recommended as an alternate therapy for ganciclovir-resistant CMV because of the frequency of ganciclovir-cidofovir cross-resistance from UL54 mutations, unless such mutations are shown to be absent and the disease is not clinically severe. There is little information on the efficacy of cidofovir in SOT.

Few novel agents for treatment of resistant CMV exist. Maribavir is an orally administered benzimidazole L-riboside that is a potent inhibitor of the CMV UL97 kinase (120,121) and while a phase II trial of maribavir as prophylaxis after stem cell transplant showed significant reduction of active CMV infection (122), phase III trials found that maribavir had similar outcomes compared to placebo (123) and a stage III trial in liver transplant recipients was halted. Maribavir has been used as salvage therapy in very limited number of cases of multidrug resistant CMV infection (124). CMX001 is a novel, orally bioavailable lipid conjugate of cidofovir that limits toxicity while providing broad-spectrum antiviral activity (including against all the human herpesviruses, as well as adenovirus and others), and which appears promising for CMV treatment. The terminase inhibitor AIC246 (Letemovir) exhibits excellent anti-HCMV activity *in vitro* and *in vivo* and currently is undergoing a clinical phase IIb trial. (125); it has been successfully used to treat resistant CMV (126). The antimalarial artesunate (127) has anti-CMV effects that may be clinically helpful, especially with resistant disease (128). Leflunomide (an immunosuppressive agent approved for rheumatoid arthritis) has antiviral activity and has been reported to clear CMV viremia in transplant recipients with resistant CMV (129–132), but failure has been reported (133).

Conclusions

CMV is the most common infection after SOT and modern methods allow for better prevention than ever before. The two main methods for CMV prevention, universal prophylaxis and preemptive therapy, decrease the risk of CMV disease, although each has its risks and benefits. Late CMV is more commonly seen in those on universal prophylaxis, and suggests CMV can be more effectively delayed than prevented, especially in primary infection (i.e. D+/R–). Preemptive therapy requires frequent testing, and very close attention to detail such that minimal viral replication occurs; there is concern that even with nominal viral replication, the indirect effects of CMV may occur. Transplant programs should choose a prophylaxis method based on local practices and experiences, including type of immunosuppression, rate of CMV seropositivity, feasibility of routine testing and costs of medication and testing. Immunologic assays are an emerging technology that will hopefully provide individualized tailoring for CMV prophylaxis and treatment.

Diagnostics has revolutionized CMV management. The recent advent of an international standard will allow for marked improvements in the ability to compare across different testing platforms.

Treatment of CMV has evolved in recent years as new data allows for increased use of oral therapy for mild-to-moderate disease. Resistant CMV is a clinical and labora-

tory diagnosis that requires careful management of antivirals, immunosuppression and adjunctive therapy.

Disclosures

The author has served as a continuing medical education speaker on the topic of CMV for Genentech and Viropharma. She led The Transplantation Society International CMV Consensus Group with an investigator-initiated independent grant from Roche, now Genentech. She has been a scientific consultant for Genentech, Roche and CSL Behring.

Box 1: Dose, formulation and duration of antiviral prophylaxis

- The dose for universal prophylaxis is half that of preemptive therapy (which is standard treatment dose). For normal renal function, the dose for universal prophylaxis is 900 mg a day.
- Doses must always be adjusted to renal function. For example, for kidney recipients with a creatinine clearance of 40–59 mL/min, the correct dose of valganciclovir for prophylaxis would be 450 mg a day. Higher doses are unlikely to convey benefit, and may result in a greater likelihood of neutropenia and side effects, in addition to higher costs. Lower doses are more likely to be associated with treatment failure and resistant virus.
- Valganciclovir is the most common agent used for prophylaxis, especially in renal transplant recipients. Optimal antiviral prophylaxis after liver transplant is controversial; while valganciclovir is approved in Europe and Canada but not approved in the United States, the majority of clinicians polled use it for prophylaxis (38). Some studies suggest high rates of failure with valganciclovir. Some experts tend to use intravenous ganciclovir (rather than valganciclovir) for the higher risk transplant recipients, such as lung or heart transplant recipients.
- For D–/R– transplants (roughly one quarter of SOT in the United States and Canada), antiviral prophylaxis against CMV is not generally necessary. Acyclovir, valacyclovir or famciclovir may be used for herpes and varicella prophylaxis.
- Duration of prophylaxis is emerging as an influential tool for decreasing the risk of CMV. Experts recommend 3–6 months of prophylaxis in D+/R– transplants (2). Some of the more convincing data are in the IMPACT trial (11), in high-risk (i.e. CMV D+/R–) kidney transplant recipients enrolled in an international, multicenter, double-blind, randomized controlled trial comparing the efficacy and safety of 200 days versus 100 days of valganciclovir prophylaxis in 326 high risk (CMV D+/R–) kidney allograft recipients. Significantly fewer patients in the

200-day group versus the 100-day group developed confirmed CMV disease by 12 months after transplant (16.1% vs. 36.8%; $p < 0.0001$); CMV viremia was also significantly lower in the 200-day group (37.4% vs. 50.9%; $p = 0.015$ at month 12).

- Optimal duration of prophylaxis in seropositive recipients remains unclear. Most experts would recommend a minimum of 3 months of prophylaxis (with either D+ or D-) in kidney, pancreas, liver, and heart transplant recipients (2). For those receiving antilymphocyte antibody induction, or lung and intestinal transplant recipients, between 3 and 6 months of prophylaxis can be used.
- Organ transplant recipients at higher risk for CMV and for complications from CMV may benefit from longer prophylaxis (2). A multicenter, placebo-controlled trial involving 11 US lung transplant centers by Palmer et al. (13) evaluated 66 lung transplant recipients (with mixed serologic combinations) who completed 3 months of valganciclovir prophylaxis and compared them with 70 who were given 12 months of prophylaxis. CMV disease occurred in 32% of the shorter course group versus 4% of the extended course group ($p < 0.001$). Significant reductions were observed with respect to severity of CMV infection, peak viremia and disease severity with extended prophylaxis. Rates of acute rejection, opportunistic infections, CMV UL97 ganciclovir-resistance mutations, adverse events and laboratory abnormalities were similar between the two groups.

Box 2: Treatment

- The most common antiviral treatment is oral valganciclovir or intravenous ganciclovir. Due to toxicities, foscarnet and cidofovir are not used as first line agents.
- For nonsevere CMV disease, oral valganciclovir (900 mg orally every 12 h) or IV ganciclovir (5 mg/kg every 12 h) are recommended as first-line treatment (93). For severe disease or retinal disease, intravenous ganciclovir is recommended due to limited trial data with oral drug in these settings.
- As with prophylaxis, optimal dosing according to renal function limits toxicity and optimizes outcomes.
- Monitoring should be performed weekly during treatment of infection. Either CMV pp65 antigenemia or CMV QNAT can be used. The use of one testing platform and one sample type (plasma or whole blood) is recommended.
- Experts recommend the “treat to negative” paradigm, or until two assays are negative a week apart. Those with higher viral loads will be slower to fall, in general, and may require longer treatment periods. Trends of serial monitoring are easier to interpret than an individual test result.

- Reduction of immunosuppression with treatment of active CMV should be made on a case-by-case basis; it should be more strongly considered in those with high viral loads, severe or slowly responsive disease, and/or in those with high “net state of immunosuppression”. Whether to resume prior levels of immunosuppression after completing treatment should be individualized as well. Recurrent CMV requires further evaluation of the overall immunosuppressive regimen, as it sometimes implies overimmunosuppression.
- Failure to respond to appropriately dosed therapy, either clinically or virologically, should trigger concerns about antiviral resistance. Clinicians may wish to test for UL97 and/or UL54 mutations to help guide further therapy, and consider switching therapy to either foscarnet, foscarnet/ganciclovir combination therapy, or high dose ganciclovir therapy, as per guidelines (2).

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Questions

- 1. The two most common methods of prevention include universal prophylaxis and preemptive therapy. Which best describes the correct hybrid approach to prevention?**
 - a. Using both antiviral prophylaxis and weekly monitoring during the first 12 weeks after transplant
 - b. Using weekly monitoring for twelve weeks after transplant, and starting prophylactic dose antiviral therapy if positive
 - c. Give patient antiviral prophylaxis for the first 3–6 months after transplant, followed by weekly monitoring for a 8–12 weeks, with plan to initiate treatment dose antiviral therapy if positive
- 2. Which of the following oral agents has the highest bioavailability AND most significant activity against cytomegalovirus?**
 - a. Acyclovir
 - b. Valganciclovir
 - c. Valacyclovir
 - d. Leflunomide
 - e. Oral ganciclovir
- 3. Diagnostically, which of the following is superior for routine monitoring?**
 - a. CMV pp65 antigenemia
 - b. CMV quantitative nucleic acid testing on plasma
 - c. CMV quantitative nucleic acid testing on whole blood
 - d. None is superior; all are effect methods
- 4. For severe, life-threatening CMV that is clinically resistant to ganciclovir, which antiviral therapy is recommended?**
 - a. Leflunomide
 - b. Sirolimus
 - c. Artesunate
 - d. Foscarnet
 - e. Ganciclovir, high doses