EDITORIAL COMMENT

Prevention of Cytomegalovirus infection and disease in Kidney Transplantation

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In transplantation of utmost importance are the interactions among immunologically active cells capable of responding to and destroying foreign tissues, the immunosuppressive therapy aimed at controlling these cells and certain infectious agents that are modulated by both factors.

Cytomegalovirus acquisition by humans (HCMV) is more common in lower socioeconomic groups. Natural routes of transmission include intrauterine, perinatal and horizontal in childhood and in the sexually active people[1]. Other routs of transmission are solid organ transplantation, blood transfusion and semen used for in vitro fertilization.

The post-transplant immunocompromised state provides an environment in which HCMV can exert its full pathogenic potential[2]. HCMV disease is a major cause of morbidity and mortality in solid organ transplantation. It is also associated to an increased risk of opportunistic infections, allograft injury and higher transplantation costs. HCMV infection also seems to increase the risk of acute and chronic rejection of the allograft via immune – mediated vascular injury[3,4]. Recent evidence suggests that influence of HCMV disease on graft survival is apparent only in patients with zero HLA-DR matches and this calls for new prophylaxis treatment and allocation strategies[5].

In solid organ transplantation the term "cytomegalovirus infection" refers to asymptomatic HCMV infection and the term "cytomegalovirus disease" refers to symptomatic HCMV infection. Disease manifestations appear most frequently between the end of the 1st and the end of the 4th month[6] after transplantation and include interstitial pneumonitis, hepatitis, gastrointestinal disease, arthritis, chorioretinitis, leukopenia and pyrexial debilitating illness, whose incidences and relative frequencies vary among transplant groups under consideration. In the absence of antiviral prophylaxis, the risk of symptomatic infection is highest in heart – lung recipients (39%) and lowest in kidney transplant recipients (8%)[7]. As many as 70 – 80% of kidney transplant recipients may show laboratory evidence of HCMV infection after renal transplantation and a significant number develop tissue invasive disease with considerable morbidity and occasional mortality[6,8]. Recently it has been suggested that within the first three months after transplantation 20% to 60% of the recipients develop HCMV disease, with a mortality rate 1% to 4%[9,10]. Therefore, prophylaxis with antiviral therapy has been recommended in patients at risk.

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Donor and recipient serostatus before transplantation

Measurement of the serostatus of donor and recipient at the time of transplant provides prognostic information about the risk of developing HCMV disease[11]. Most of the renal transplant recipients (>90%) are seropositive before transplantation. These patients should be regarded as possessing latent HCMV capable of reactivation when become immunocompromized because of immunosuppression. In these cases HCMV may reactivate from latency and may cause disease. Also, organs harvested from seropositive for HCMV individuals for transplantation may transmit virus, irrespective of whether the recipient is seronegative or seropositive. It has been observed that recipients with HCMV seropositive and zero HLA – DR matched kidneys are at a considerably increased risk of HCMV associated graft loss[5]. Seropositive recipients frequently excrete HCMV in the urine but have a low risk of HCMV

disease unless the donor is seropositive[12]. The risk for HCMV disease is highest in donor – seropositive and solid organ transplant recipient – seronegative (D+/R-). In spite of this, many raise the question whether prophylaxis with antiviral therapy should be recommended for all seronegative patients on conventional immunosuppression who receive a kidney from a seropositive donor. In seronegative patients receiving a seronegative kidney and in seropositive recipients, the risk of HCMV disease is low. Therefore, no antiviral therapy is required.

Immunosuppression

The recipient's overall status of immunosuppression determined by the immunosuppressive protocol (e.g. type of the drug, dose, timing, duration), age, primary renal disease, co-morbidity is another risk factor. Increasing immunosuppression to lower the risk of acute rejection or to treat episodes of acute rejection increases the incidence of infections and malignancies[13,14].

Regarding the type of immunosuppression, antilymphocyte antibodies (ALA) and monoclonal antibodies (such as OKT3) as either induction or antirejection therapy cause a high incidence of HCMV disease. The risk is maximal when ALA therapy is used for the treatment of organ rejection, with HCMV disease being diagnosed three to four times more frequently than in patients not receiving antilymphocyte antibody therapy[15]. Transplant recipients in whom primary infection develops at the time of transplantation (donor: cytomegalovirus antibody positive; recipient: cytomegalovirus antibody negative) or who test HCMV antibody positive before transplantation and require antilymphocyte antibody therapy after transplantation have a greater than 50% attack rate of cytomegalovirus disease[16]. There is agreement that all recipients treated with antilymphocyte antibodies should receive prophylaxis independently of recipient and donor seropositivity[17]. The use of interleukin – 2 receptor monoclonal antibodies does not seem to augment the incidence of HCMV infection[18] but a long-term follow up period is crucial to draw firm conclusions. Cyclosporine, tacrolimus, rapamycin and prednisone do not activate latent virus but once replicating virus is present these drugs potent amplification. Azathioprine and cyclophosphamide have moderate effect on both reactivation and replication. Mycophenolate mofetil has been suggested to increase the incidence of HCMV infection compared with azathioprine regimens[19].

Other risk factors

Kidney transplant recipients appear to be at increased risk of HCMV infection and disease when there is zero HLA matching[20] and more recently when there are zero HLA – DR matches and seropositive donors[5].

In the steady state of a latent HCMV infection, HCMV-specific CD4+ T cells dominate the immune response. A loss of these cells in the first months after transplantation of HCMV seropositive renal transplant recipients not only correlates with an uncontrolled viral replication but also with an increased incidence of HCMV-related disease[21]. In contrast, during primary HCMV infection, the cellular immune response is dominated by HCMV – specific CD8+ T cells[22].

It has been shown that peak HCMV load during active infection is a major risk factor that correlates with the development of HCMV disease. Also, it has been shown that the classical risk factors of donor / recipient serostatus were entirely explained by viral load[23-26].

Data from the USRDS database showed an increasing number of rejections due to infections in older recipients, whereas the number of rejections decreased with age[27,28].

Management of HCMV

Prevention of infections is one of the primary goals in the management of renal transplant recipient. Theoretically the best chance of prevention is represented by low immunosuppression. However, avoiding rejection, without exposing the patient to infection or re – infection, remains a difficult task even with modern immunosuppressive therapy. A number of measures may be taken to prevent infections.

The principles of managing HCMV infection and disease in the immunocompromised host are to anticipate their development, to define policies for monitoring patients routinely for the presence of viremia according to their baseline risk of HCMV disease and to enhance surveillance if patients develop a condition likely to increase their risk of HCMV disease. The patients then will be offered prophylaxis or preemptive therapy based on an assessment of their individualized risk of disease, together with data from controlled clinical trials in the same patient group supporting the efficacy and safety of possible antiviral interventions.

Prophylaxis and preemptive therapy are strategies used for managing patients at risk for HCMV disease. Each therapy has advantages and disadvan-

tages that must be considered in the context of the patient and the allograft as well as the laboratory facilities at any time point. Preemptive therapy requires a rapid and sensitive assay to detect HCMV infection sufficiently in advance of symptoms. Preemptive therapy avoids unnecessary exposure to a drug in patients who are not at increased risk of HCMV disease or HCMV – associated death.

Several prophylactic, preemptive and deferred therapeutic regimens are in use, with variable success[9,10,29]. Deferred therapy (started when overt HCMV disease manifestations are present) was shown to be inferior to prophylaxis with oral ganciclovir[30]. Prophylaxis with ganciclovir, which is the reference treatment for HCMV disease, frequently results in unnecessary, potentially harmful treatment. Furthermore, long – term antiviral treatment may expose some patients to drug toxicity, requires maintenance access, is expensive and may lead to generation of resistant viruses. The problem of intravenus access should be resolved with oral valganciclovir (Valcyte®), which seems as efficient as intravenous ganciclovir.

True and delayed prophylaxis

We say *true prophylaxis* when the drug is administered from the first day of transplantation before there is any active viral replication. This strategy may be used where an assessment at baseline shows that the risk of disease is high, the chance of severe disease is also high, and at least one double – blind, randomized, pladebo – controlled trial supports the efficacy and safety of prophylaxis in the target population. The patient then will be given the drug from the time of transplant onward and continued for the duration studied in the controlled clinical trial that provided evidence for its use. Immunoglobulins, acyclovir, ganciclovir, valaciclovir and valganciclovir have been used for prevention of HCMV infection in organ transplant patients. In *delayed prophylaxis*, at baseline a decision was made that true prophylaxis was not necessary. However the patient's immunological status for some reason has changed and is decided that prophylaxis therapy must start now.

The major disadvantage of true prophylaxis is that a large number of patients that is not going to develop HCMV disease will receive unnecessary therapy. This, together with the cost, the risk for side effects of the antiviral compound (ganciclovir toxicity)[31] and the possibility of emergence of resistant HCMV strains[31] implies that it is not an ideal approach. Also, prophylactic therapy can not completely prevent primary infection of the recipient[32,33].

Among seronegative patients undergoing seroconversion, the median time until viral DNA was detectable appears to be approximately 25 to 30 days[34]. This time was similar in patients who received three month true prophylaxis. Thus prophylaxis reduces the incidence of primary infection and delays the event of it to a time period of less intense immunosuppression and improved immunocompetence. We already know that the prolongation of prophylaxis treatment is beneficial in some cases[5].

Acyclovir (Zovirax®) in a dose of 800-3200 mg per day depending on renal function, for 12 weeks, reduced the incidence of HCMV infection and disease after kidney transplantation in a trial by Balfour and colleagues[35]. The most striking benefit of high – dose acyclovir was the prevention of acquisition of primary HCMV disease from a seropositive donor . Moreover, acyclovir had an effect on the incidence of HCMV infection and disease in seropositive recipients.

True or delayed prophylaxis with ganciclovir in renal transplant recipients, HCMV antibody positive before transplantation, during antilymphocyte serum administration showed a significant reduction in the incidence of HCMV disease[36]. Intravenous ganciclovir is given at doses ranging between 5 and 10 mg/kg/d for 10 – 14 days. Oral ganciclovir is given at doses of 1.5 – 2.0 g / d for 14 – 90 days. A recent meta analysis of 13 controlled trials showed that prophylactic treatment with acyclovir or ganciclovir could significantly reduce the risk of HCMV disease and infection, with no difference between the two agents[37].

High dose of oral acyclovir is not as effective as oral ganciclovir in preventing HCMV infection and disease in kidney transplant recipients who have received muromonab CD3 for induction[38]. In these high-risk patients, acyclovir for three months provided effective prophylaxis only for recipients of seronegative donor kidneys. In contrast ganciclovir given for three months was an effective prophylactic agent for recipients of seropositive donor kidneys.

Valaciclovir, 2 g four times per day, adjusted for renal function, administered for three months posttransplantation reduced the incidence of, and increased the time to, HCMV disease in seropositive donor and seronegative kidney transplant recipients. At the end of treatment period, 45% of seronegative receiving placebo recipients had confirmed HCMV disease versus 3% of seronegative patients receiving valaciclovir. In seropositive pa-

tients, the incidence of HCMV disease was 6% in those receiving placebo and 0% in valaciclovir recipients[32]. Prophylaxis for HCMV with both valaciclovir and intravenous ganciclovir was less costly and at least as effective as preemptive therapy, adjusting the immunosuppression and wait – and – treat strategies[39]. Routine HCMV pp65 antigenemia monitoring shows that some patients develop positive pp65 antigenemia during valaciclovir treatment or after prophylaxis cessation. These cases can be efficiently treated by reduction of immunosuppression therapy without immunologic complication. This strategy may prevent abusive use of IV ganciclovir and eventually the occurrence of HCMV resistance to this drug[40]. At least one multicentric study has confirmed the beneficial effect of valaciclovir in reducing and delaying the onset of HCMV disease after kidney, heart and bone marrow transplantation[41]. However, the report of cases of haemolytic uremic syndrom in HIV – positive patients taking valaciclovir has made its use questionable[42].

A significant reduction of the incidence of HCMV infection and disease has been obtained with intravenous immunoglobulins[10]. A drawback of immunoglobulins is that treatment should be prolonged for at least 16 weeks[17]. Other disadvantages of using immunoglobulins are the cost of treatment and the heterogeneity of the preparation.

HCMV – seropositive patients were associated with a significantly higher incidence of delayed graft function, lower rates of HCMV – disease, a lower incidence of graft loss and lower medicare costs than seronegative recipients. HCMV - seronegative recipients with HCMV – seropositive donors compared to seronegative recipients with seronegative donors were associated with significantly higher incidence of HCMV disease, graft loss, and higher costs. Donor serology has a larger impact than recipient serology and the donor effects are more pronounced in HCMV – seronegative recipients[43].

Suppression and preemptive therapy

In *suppression therapy* the patient has been monitored weekly for viral replication and the measurements have shown that there is moderate predictive positive value for HCMV disease (for example 30%). Then, an antiviral agent is given with the intension to suppress viral replication below the level needed to cause viremia. Suppression requires a drug more potent than that needed for prophylaxis.

The term *preemptive therapy* describes intervention when the results of laboratory tests indicate that the patient is at imminent risk of HCMV disease. It is used after detection of viremia in any immunocompromised patient. In this case the patient has been monitored by collecting weekly samples of blood processed by laboratory methods known to provide a high positive predictive value for HCMV disease (e.g. 50 – 60%). In this case it is decided to give an antiviral agent with the intention of halting HCMV viremia before it reaches the high viral loads required to cause disease (Table 1).

Table 1. *Guidelines to prophylaxis and preemptive therapy*

Antiviral prophylaxis
Donor$^+$/Recipient$^-$
ALA induction therapy
Rejection therapy with ALA
Preemptive therapy
When significant virus replication before onset of symptoms
Concomitant lowering of immunosuppression

Decision points for starting preemptive therapy must be based on the results of clinicopathologic studies with the assay under evaluation. Antigenemia recorded by pp65 antigen test is closely correlated with symptomatic HCMV infection. This rapid diagnostic method has replaced cell culture procedures to a large extent. Examples include detection of viremia by PCR or antigenemia above a cutoff value associated with a high risk of disease[44] or two consecutive samples HCMV PCR-positive. In the study of Koetz[44] preemptive therapy was started in the high-risk patients (D+/R-) at the first pp65 antigen – positive test result. In contrast, to prevent HCMV – associated symptoms in pretransplantation seropositive patients, it was sufficient to start preemptive therapy at pp65 – antigen levels of >5 positive cells /2X10^5. More recently, the results of viral dynamic assessments have been applied to this problem. Patients at risk of disease can be identified by the absolute value of viral load found in the first HCMV PCR-positive sample, coupled with an assessment of individual viral dynamics by calculating the rate of increase from the last HCMV PCR-negative sample.

The existing data suggest that ganciclovir reduces the viral load during the primary viremia and indirectly assists both in rapid induction and maintenance of HCMV – specific cellular immune responses. After acute HCMV infection the decrease in viral load and the reappearance or stabilization of HCMV –

specific CD4+ T cells may serve as a combined parameter to define the time point at which therapeutic antiviral treatment can be withdrawn or preemptive monitoring of HCMV load may be stopped[22].

While drug costs for antiviral prophylactic therapy are not insignificant, the alternative approach, preemptive therapy to prevent HCMV disease does not eliminate HCMV disease and HCMV viremia may recur in up to 25% of patients[10]. HCMV treatment after the detection of HCMV infection necessitates the use of more potent antiviral agents. The only agent currently licensed for this indication is intravenous ganciclovir which is associated with a significant risk of neutropenia and increased need for patient hospitalization. Underdosing of ganciclovir can lead to development of resistant HCMV populations. Foscarnet and cidofovir are alternative options for patients with resistance to ganciclovir but their use in transplantation is of limited value because of their nephrotoxicity. Valganciclovir, an orally administered prodrug of ganciclovir (Table 2) has excellent bioavailability and achieves serum levels similar to intravenous ganciclovir[45,46]. Valganciclovir has recently shown similar efficacy as i.v. ganciclovir in the treatment of HCMV chorioretinitis in HIV – positive individuals[47].

The preemptive therapy must be accompanied by reduction of immunosuppressive therapy until documented control of HCMV infection. The decrease or discontinuation of MMF is suggested. Calcineurin inhibitors are maintained at therapeutic levels except of cases with interstitial pneumonitis in which they must be reduced or stopped. In these cases steroids are given in a dose of 16 mg /d methylprednizolone or equivalent.

Prophylaxis of the fetus

Because pregnancy is recommended two years after successful transplantation, HCMV infection in pregnant renal transplant recipients seems to be low. At 2 years, the risk of HCMV infection is less than in the first post – transplant year. However, primary HCMV infection or reactivation can be transmitted to the fetus[48]. Congenital HCMV infection is associated with perinatal death, microcephalus or mental retardation in 10% of cases, and some children apparently normal at delivery, may show hearing loss and learning problems later on. Culturing of amniotic fluid is essential to allow HCMV diagnosis in the fetus[48]. The efficacy and safety of treating the mother with ganciclovir or HCMV hyperimmunoglobulin to prevent fetal HCMV disease are not known. This therapy should be only started after considering the safety of the mother, as ganciclovir can induce birth defects in animals. Hou recommended measuring titers of anti – HCMV IgG and IgM every three months during pregnancy[49].

A center without access to molecular methods or pp65 antigenemia assay should use prophylaxis rather than pre – emptive therapy for HCMV disease prevention.

Table 2. ***Dose adjustment of valganciclovir according to renal function***

Creatinine clearance Ml/min	Starting dose mg	Maintenance dose mg
> 60	2x900	1x900
40-59	2x450	1x450
25-39	1x450	450 oad
10-24	450 oad	450 tw
haemodialysis	450 following dialysis sessions	

oad: on other days, tw: twice weekly

Methods used for viremia detection

Early initiation of antiviral therapy offers the greatest benefit for the patient. Hence, regular screening of transplant patients, especially during the critical three – month posttransplant period, for the presence of HCMV in blood and other specimens is of utmost importance for effective patient management.

Increasing levels of HCMV load in the leukocytes or in the serum precede overt HCMV disease. Thus primary HCMV infection can be diagnosed at the beginning and HCMV reactivation can be assessed quantifying HCMV load. Pre – emptive therapy is based on quantifying systemic viral load and initiating therapy at a given threshold. Serological diagnosis of HCMV infection in the immunocompromised is both insensitive and subject to distortion by passive antibody administration. Sensitive tests are now available to diagnose HCMV infection or disease and hence lead a) to the initiation of treatment, b) to monitor antiviral treatment strategies c) to determine treatment failures.

Assays using leukocytes are more sensitive and more rapidly positive after transplantation than those using plasma or serum. The tests for detection of HCMV infection fall into three broad categories: molecular assays, non – molecular assays and other assays (Table 3).

Table 3. ***Assays for the detection of HCMV infection***

Molecular assays	Non – Molecular assays	Other assays
CMV-DNA Quantitative PCR assay	Conventional cell culture	Serology
CMV-DNA Qualitative PCR assay	Shell vial assay	Histopathology
NASBA CMV pp67 assay	pp65 antigenemia assay	
Hybrid Capture CMV DNA assay		
Branched - DNA (bDNA) assay		

There are some prerequisites for an assay for guiding pre – emptive therapy. The most important are a) high sensitivity that allows early detection of HCMV infection in patients at high risk of disease, b) the potential to quantify the results, to increase the positive predictive value and to measure viral load during treatment, c) to allow early initiation or change of treatment rapidly and d) to have high degree of reproducibility[50].

The low sensitivity and low reproducibility of conventional cell culture and the shell vial assay limit their role in the management of HCMV infection to the one of diagnosis of disease. Even for this purpose, the turnaround time of the assay, which can be up to 4 weeks for conventional cell culture, must be considered. Newer diagnostic assays (for example pp65 antigenemia assay and molecular assays) have improved the ability to diagnose HCMV quickly and accurately. These methods fulfill most of the requirements for a diagnostic assay: they have high sensitivity, they can quantify viral load, and they are rapid and reproducible. Their characteristics should allow the assays to be used to predict the development of HCMV disease and monitor response to therapy.

Weekly monitoring of HCMV viremia using pp65 antigenemia assay or a molecular method during the first 3 months after transplantation or longer during intensification of immunosuppression is recommended. Although molecular methods are more sensitive than the pp65 antigenemia assay, the choice of test must consider availability and cost. The detection of pp65 is accomplished by using indirect immunofluorescence[51]. A threshold of = 10 positive cells/ $2x10^5$ cells is required to guide pre – emptive therapy for solid organ transplant recipients[52,53].

References

1. Weller TH. The cytomegaloviruses: ubiquitous agents with protean clinical manifestations. N Engl J Med 1971; 285:203-214
2. Rubin RH. The indirect effects of cytomegalovirus infection on the outcome of organ transplantation. JAMA 1989; 261:3607–3609
3. Valantine HA. Prevention and treatment of cytomegalovirus disease in thoracic organ transplant patients: evidence for a beneficial effect of hyperimmune globulin. Transplant Proc 1995; 27(Suppl 1):49-57
4. Goral S, Helderman JH. Cytomegalovirus and rejection. Transplant Proc 1994; 26:5
5. Schnitzler MA, Lowell JA, Hmiel SP, et al. Cytomegalovirus disease after prophylaxis with oral ganciclovir in renal transplantation: The importance of HLA-DR matching. J Am S Nephrol 2003; 780-785
6. Rubin RH. Infectious disease complications of renal transplantation. Kidney Int 1993; 44: 221 – 236
7. Ho M. Advances in understanding cytomegalovirus infection after transplantation. Transplant Proc 1994; 26:7-11
8. Murray BM, Amsterdam D, Gray V, et al. Monitoring and diagnosis of cytomegalovirus infection in renal transplantation. J Am Soc Nephrol 1997; 8:1448-1457
9. Brennan DC, Garlock KA, Lippmann BA, et al. Control of cytomegalovirus – associated morbidity in renal transplant patients using intensive monitoring and either preemptive or deferred therapy. J Am Soc Nephrol 1997; 8:118-125
10. Patel R, Snydman DR, Rubin RH, et al. Cytomegalovirus prophylaxis in solid organ transplant recipients. Transplantation 1996; 61:1279-1289
11. Betts RF, Freeman RB, Douglas RJ Jr, and Talley TE. Clinical manifestations of renal allograft derived primary cytomegalovirus infection. Am J Dis Child 1977; 131:759-763
12. Chou SW. Reactivation and recombination of multiple cytomegalovirus strains from individual organ donors. J Infect Dis 1989; 160:11-15
13. Kamel O, van de Rijn M, Lebrun D, Weiss L, Warnke R, Dorfman R. A multivariate analysis of discharge immunosuppression and posttransplant malignancy. Am J Transplant 2001; 1(Suppl 1):353
14. Jamil B, Nickills K, Becker G, Walker R. Impact of acute rejection therapy on infections and malignancies in renal transplant recipients. Transplantation 1999; 68:1597-1619

15. Sia IG, Patel R. New strategies for prevention and therapy of cytomegalovirus infection and disease in solid – organ transplant recipients. Clin Nephrol Rev 2000; 13:83-121
16. Hibberd PI, Tolkoff – Rubin NE, Cosimi AB, et al. Symptomatic CMV disease in the CMV antibody seropositive renal transplant recipient treated with OKT3. Transplantation 1992;53:68-72
17. Jassal SV, Roscoe JM, Zaltzman JS, et al. Clinical practice guidelines: prevention of cytomegalovirus disease after renal transplantation. J Am Soc Nephrol 1998; 9:1697-1708
18. Adu D, Cockwell P, Ives N, Shaw J, Wheatley K. Interleukin – 2 receptor monoclonal antibodies in renal transplantation: meta – analysis of randomized trials. Br Med J 2003; 326:789-793
19. Sollinger HW. Mycophenolate mofetil for the prevention of acute rejection in primary cadaveric renal allograft recipients. U.S. renal transplant mycophenolate mofetil study group. Transplantation 1995; 60:225-232
20. Fishman JA. Prevention of CMV infection in transplant patients. Transplant Infect Dis 1999; 1:35-39
21. 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; 71:1287-1294
22. Sester MS, Sester U, Gartner BC, Girndt M, Meyerhans A, Kohler H. Dominance of virus – specific CD8+ T cells in human primary cytomegalovirus infection. J Am Soc Nephrol 2002; 13:2577-2584
23. Cope AV, Sabin C, Burroughs A, Rolles K, Griffiths PD, Emery VC. Interrelationships among quantity of human cytomegalovirus DNA in blood, donor – recipient serostatus and administration of methylprednisolone as risk factor for HCMV disease following liver transplantation. J Infect Dis 1997; 176:1484-1490
24. Cope AV, Sweny P, Savin C, Rees L, Griffiths PD, Emery VC. Quantity of cytomegalovirus viruria as a major risk factor for cytomegalovirus disease after renal transplantation. J Med Virol 1997; 52:200-205
25. Gor D, Sabin C, Prentice HG, et al. Longitudinal fluctuations between peak virus load, donor/recipient serostatus, acute GvHD and CMV disease. Bone Marrow Transplant 1998; 21:597-605
26. Hassan – Walker AF, Kidd JM, Sabin C, Sweny P, Griffith PD, Emery VC. Quantity of human cytomegalovirus DNAemia as a risk factor for CMV disease in renal allograft recipients: relationship with donor recipient serostatus, recipient of augmented methylprednisolone and anti-thymocyte globulin. J Med Virol 1999; 58:182-187
27. Meier – Kriesche HU, Friedman G, Jacobs M, et al. Infectious complications in geriatric renal transplant patients: comparison of two immunosuppressive protocols. Transplantation 1999; 68:1496-1502
28. Meier – Kriesche HU, Ojo AO, Arndorfer JA, et al. Need for individualized immunosuppression in elderly renal transplant recipients. Transplant Proc 2001; 33:1190-1191
29. Griffiths PD. Prophylaxis against CMV infection in transplant patients. J Antimicrob Chemother 1997; 39:299-301
30. Brennan DC, Burton KG, Singer GG, et al. Prophylactic oral ganciclovir compared with deferred therapy for control of cytomegalovirus in renal transplant recipients. Transplantation 1997; 64:1843-1846
31. Limaye AP, Corey L, Koelle DM, Davis CL, Boeckh M. Emergence of ganciclovir resistant cytomegalovirus disease among recipients of solid – organ transplants [See comments]. Lancet 2000; 356: 645-649
32. Lowance D, Neumayer HH, Legendre CM, et al. Valacyclovir for the prevention of cytomegalovirus disease after renal transplantation. N Engl J Med 1999; 340:1462-1470
33. Razonable RR, Rivero A, Rondriguez A, et al. Allograft rejection predicts the occurrence of late – onset cytomegalovirus disease among CMV – mismatched solid organ transplant patients receiving prophylaxis with oral ganciclovir. J Infect Dis 2001; 184:1461-1464
34. Rentenaar RJ, Gamadia LE, van DerHoek N, et al. Development of virus – specific CD4+ T cells during primary cytomegalovirus infection. J Clin Invest 2000; 105:541-548
35. Balfour HHJr, Chase BA, Stapleton JT, Simmons RL, Fryd DS. A randomized, placebo – controlled trial of oral acyclovir for the prevention of cytomegalovirus disease in recipients of renal allografts. N Engl J Med 1989; 320:1381-1387
36. Hibberd PI, Tolkoff – Rubin NE, Conti D, et al. Preemptive ganciclovir therapy to prevent cytomegalovirus disease in cytomegalovirus antiboby – positive renal transplant recipients. Ann Int Med 1995; 123:18-26
37. Couchoud C, Cucherat M, Haugh M, Poyteil – Noble C. Cytomegalovirus prophylaxis with antiviral agents in solid organ transplantation. Transplantation 1998; 65:641-647
38. Flechner SM, Avery RK, Fisher R, et al. A randomized prospective controlled trial of oral acyclovir versus oral ganciclovir for cytomegalovirus prophylaxis in high – risk kidney transplant recipients. Transplantation 1998; 66:1682-1688
39. Mauskopf JA, Richter A, Annemans L, Maclaine G. Cost – effectiveness model of cytomegalovirus management strategies in renal transplantation. Comparing valaciclovir prophylaxis with current practice. Pharmacoeconomics 2000; 18:239-251
40. Anglicheau D, Lautrette A, Scieux C, et al. Lowering immunosuppression is safe and effective to treat altered pattern of CMV infection in renal transplant recipients on valaciclovir prophylaxis. Transplant

Proc 2000; 34:2826-2827
41. Squifflet JP, Kovarik J, Brennan PJ, et al. Valaciclovir for the prevention of cytomegalovirus disease after renal transplantation. International Valacyclovir Cytomegalovirus prophylaxis Transplantation Study Group. N Engl J Med 1999; 340:1462-1470
42. Bell WR, Chulay JD, Feinberg JE. Manifestations resembling thrombotic microangiopathy in patients with advanced human immunodeficiency virus (HIV) disease in a cytomegalovirus prophylaxis trial. Medicine 1997; 76:369-380
43. Schnitzler MA, Lowell JA, Hardinger KL, Boxerman SB, Bailey TC, Brennan DC. The association of cytomegalovirus sero – pairing with outcomes and costs following cadaveric renal transplantation prior to the introduction of oral ganciclovir CMV prophylaxis. Am J Transplant 2003; 3:445-451
44. Koetz AC, Delbruck R, Furtwangler A, et al. Cytomegalovirus pp65 antigen – guided preemptive therapy with ganciclovir in solid organ transplant recipients: A prospective double – blind, placebo – controlled study. Transplantation 2001; 72:1325-1327
45. Pescovitz MD, Rabkin J, Merion RM, et al. Valganciclovir results in improved oral absorption of ganciclovir in liver transplant recipients. Antimicrob Agents Chemother 2000; 44:2811-2815
46. Czock D, Scholle C, Rasche FM, Schaarschmidt D, Keller F. Pharmacokinetics of valganciclovir and ganciclovir in renal impairment. Clin Pharmacol Ther 2002;72:142-150
47. Martin DF, Sierra – Madero J, Walmsley S, et al. A controlled trial of valganciclovir as induction therapy for cytomegalovirus retinitis. N Engl J Med 2002; 346:119-126
48. Hagay ZJ, Biran G, Ornoy A, Reece EA. Congenital cytomegalovirus infection: a long – standing problem still seeking a solution. Am J Obstet Gynecol 1997; 174:241-245
49. Vergoulas G. The reproductive function after renal transplantation. In Vergoulas G (ed), "Kidney Transplantation", Parisianos, Thessaloniki – Athens, 2000, pp 353-374
50. Boeckh M, Boivin G. Quantitation of cytomegalovirus methodologic aspects and clinical applications. Clin Microbiol Rev 1998; 11: 533-554
51. Meyer – Koning U, Serr A, Hufert FT, et al. Laboratory diagnosis of HCMV – related disease in renal transplant patients – pp65 antigen detection versus nested PCR. Clin Diagn Virol 1995; 3:49-53
52. van den Berg AP, van der Bij W, van Son WL, et al. Cytomegalovirus antigenemia as a useful marker of symptomatic cytomegalovirus infection after renal transplantation – a report of 130 consecutive patients. Transplantation 1989; 48:991- 995
53. Halwachs G, Zach R, Pogglitsch H, et al. A rapid immunocytochemical assay for HCMV detection in peripheral blood of organ – transplanted patients in clinical practice. Transplantation 1993; 56: 19-24

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