OPEN

Therapeutic Drug Monitoring of Ganciclovir: Where Are We?

Anne-Grete Märtson, MSc, Angela E. Edwina, MSc,* Hannah Yejin Kim, PhD,†‡§ Marjolein Knoester, PhD,¶ Daan J. Touw, PhD,*‖ Marieke G. G. Sturkenboom, PhD,* and Jan-Willem C. Alffenaar, PhD†‡§*

Background: Ganciclovir is the mainstay of therapy for the prophylaxis and treatment of *Cytomegalovirus*. However, therapy with this antiviral agent is hindered by side effects such as myelosuppression, which often leads to therapy cessation. Underdosing, as an attempt to prevent side effects, can lead to drug resistance and therapy failure. Therapeutic drug monitoring (TDM) has been used to overcome these problems. The purpose of this narrative review was to give an overview of ganciclovir TDM, available assays, population pharmacokinetic models, and discuss the current knowledge gaps.

Methods: For this narrative review, a nonsystematic literature search was performed on the PubMed database in April 2021. The following search terms were used: ganciclovir, valganciclovir, pharmacokinetics, pharmacodynamics, population pharmacokinetics, therapeutic drug monitoring, bioassay, liquid chromatography coupled with tandem mass spectrometry, liquid chromatography, chromatography, spectrophotometry, and toxicity. In addition, the reference lists of the included articles were screened.

Results: The most common bioanalysis method identified was liquid chromatography coupled with tandem mass spectrometry. There are different models presenting ganciclovir IC_{50}; however, establishing a pharmacokinetic/pharmacodynamic target for ganciclovir based on preclinical data is difficult because there are no studies combining dynamic drug exposure in relation to inhibition of viral replication. The data on ganciclovir TDM show large interindividual variability, indicating that TDM may play a role in modifying the dose to reduce toxicity and prevent treatment failure related to low concentrations. The main hurdle for implementing TDM is the lack of robust data to define a therapeutic window.

Conclusions: Although the pharmacokinetics (PK) involved is relatively well-described, both the pharmacodynamics (PD) and pharmacokinetic/pharmacodynamic relationship are not. This is because the studies conducted to date have mainly focused on estimating ganciclovir exposure, and owing to the limited therapeutic options for CMV infections, future studies on ganciclovir are warranted.

Key Words: ganciclovir, valganciclovir, therapeutic drug monitoring, cytomegalovirus

(*Ther Drug Monit* 2022;44:138–147)

INTRODUCTION

Cytomegalovirus (CMV) is a major complication in immunocompromised patients, particularly in hematopoietic stem cell transplant (HSCT) and solid-organ transplant (SOT) recipients.[1] Ganciclovir is the mainstay of therapy for the prophylaxis and treatment of CMV in SOT recipients.[2,3] Ganciclovir, or 9-(1,3-dihydroxy-2-propoxymethyl)guanine, is a cyclic analog of the endogenous purine nucleoside guanosine.[4] Ganciclovir is administered intravenously, whereas the prodrug valganciclovir is administered orally and gets hydrolyzed to ganciclovir postabsorption (bioavailability of a single dose of valganciclovir is approximately 60%[5]). The antiviral activity of ganciclovir requires intracellular phosphorylation and activation by the UL97 viral kinase and UL54 DNA polymerase. Ganciclovir monophosphate is further phosphorylated to ganciclovir triphosphate by cellular kinases and inhibits CMV DNA polymerase[6–10] (Fig. 1). Once ganciclovir triphosphate is formed, it seems to be very stable and persists in CMV-infected cells for several days, with an intracellular half-life ($t_{1/2}$) of 16.5 hours.[6]

For the treatment of CMV in both SOT and HSCT, intravenous (IV) ganciclovir at a dose of i.v. 5 mg/kg or oral

Received for publication June 30, 2021; accepted August 7, 2021.

From the *Department of Clinical Pharmacy and Pharmacology, University of Groningen, University Medical Center Groningen, Groningen, the Netherlands; †Sydney Pharmacy School, Faculty of Medicine and Health, The University of Sydney, Camperdown, NSW, Australia; ‡Department of Pharmacy, Westmead Hospital, Westmead, NSW, Australia; §Marie Bashir Institute of Infectious Diseases and Biosecurity, The University of Sydney, Camperdown, NSW, Australia; ¶Department of Medical Microbiology and Infection Prevention, University of Groningen, University Medical Center Groningen, Groningen, the Netherlands; and ‖Department of Pharmaceutical Analysis, Groningen Research Institute of Pharmacy, University of Groningen, University Medical Center Groningen, Groningen, the Netherlands.

A.-G. Märtson was funded by the Marie Skłodowska-Curie Actions (grant agreement no. 713660, PRONKJEWAIL—H2020-MSCA-COFUND, 2015). A. E. Edwina was funded by the Indonesian Endowment Fund for Education.

None of the authors has any conflicting interests to disclose.

Correspondence: Anne-Grete Märtson, MSc, Department of Clinical Pharmacy and Pharmacology, University of Groningen, University Medical Center Groningen, P.O. Box 30.001 (internal post code AP50) 9700 RB Groningen, the Netherlands (e-mail: a.martson@umcg.nl).

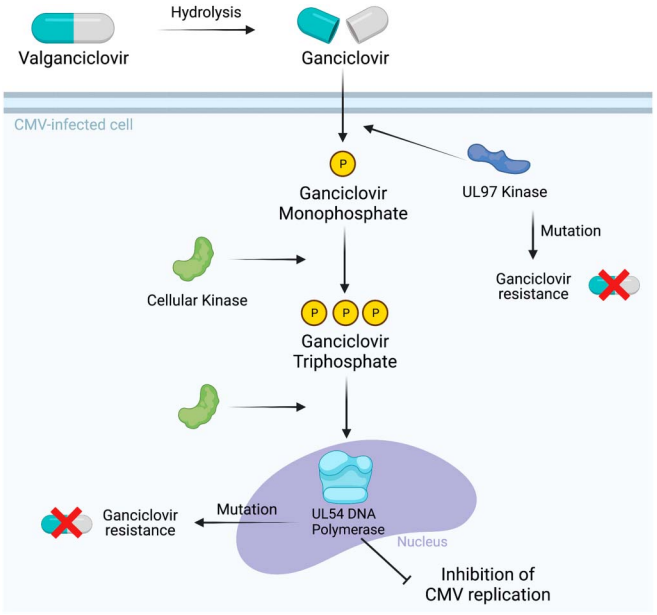

FIGURE 1. Antiviral mechanism of ganciclovir.

valganciclovir at a dose of p.o. 900 mg twice daily is the recommended regimens in adults with normal renal function.[2] Viral load thresholds at which the treatment is started may differ depending on the risk profile and immune status of the patients.[11,12] To prevent CMV infection or reactivation after SOT, prophylaxis is recommended depending on the donor and recipient CMV IgG status and the transplanted organ.[2,12,13] Regarding donor and recipient CMV IgG status, the risk of CMV complications is, for example, highest if the recipient is seronegative and the donor is seropositive and lowest when both are negative.[1,2,12,13] Transplants for which higher immunosuppressive regimens are needed (eg, lung transplantation) pose a higher risk of CMV reactivation than those that require less immunosuppression (eg, liver transplantation).[1,2,12,13] Duration of prophylaxis may range from 3 to 12 months.[2,13]

Ganciclovir toxicity can cause myelosuppression, that is, neutropenia, thrombocytopenia, and leukopenia, which can lead to dosage changes or cessation of therapy.[14–16] The rate of myelotoxicity varies between the specific patient groups. In HSCT, the rates seem to be 50% and higher, whereas in SOT, much lower rates of approximately 10% have been reported.[16–18] Therefore, pre-emptive treatment is mostly used after HSCT to avoid the side effects of (val)ganciclovir. In pre-emptive treatment, the treatment is initiated when CMV is detected by routine monitoring but before the onset of symptoms. Granulocyte colony–stimulating factor has been used to manage the myelotoxicity caused by ganciclovir.[19,20] However, the occurrence of myelotoxicity can lead to a clinician-directed dose reduction. By contrast, ganciclovir underexposure can lead to viral drug resistance, which is caused by mutations in the *UL97* and *UL54* genes[21–24] (Fig. 1).

Ganciclovir toxicity and acquired drug resistance have led to the implementation of therapeutic drug monitoring (TDM) in different centers. Although the benefits of TDM are still being debated on, there is a lack of clear targets for therapy optimization. In this review, we will provide an overview of the available bioanalytical methods and data on preclinical and clinical pharmacokinetics (PK) and pharmacodynamics (PD), which are being used for the estimation of ganciclovir exposure and as evidence for the benefits of TDM of ganciclovir.

METHODS

For this narrative review, a nonsystematic literature search was performed on the PubMed database in April 2021. The following search terms were used: ganciclovir, valganciclovir, pharmacokinetics, pharmacodynamics, population pharmacokinetics, therapeutic drug monitoring, bioassay, LC-MS/MS, liquid chromatography, chromatography, spectrophotometry, and toxicity. In addition, the reference lists of the included articles were screened.

BIOANALYSIS

A number of assay procedures using high-performance liquid chromatography and ultra-high–performance liquid chromatography (UPLC) with detectors, such as mass spectrometry, fluorescence spectrophotometry, diode array detectors, UV spectrophotometry, and pulsed amperometers, have been developed to quantify ganciclovir concentrations in biological matrices, especially serum and plasma.[25–31] A total of 14 liquid chromatographic methods with various detection methods are summarized in Table 1. Six studies described the development of liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS).[32–37] One study used capillary electrophoretic methodology,[38] whereas another used Raman spectroscopy to detect ganciclovir after ocular administration.[39]

LC-MS/MS is the preferred method to quantify ganciclovir concentrations because of its high sensitivity, selectivity, and simple sample pretreatment, especially when stable isotopes of ganciclovir are used as an internal standard.[36] The ideal assay should have the ability to quantify ganciclovir concentrations in the concentration range that can be expected in patients [from the trough (C_{min}) to the peak (C_{max}) concentration].

According to the summarized studies, run times of ganciclovir assays ranged from 2.5 to 15 minutes. Shorter run times are desirable for the efficient use of the equipment. The LC-MS/MS method developed by Singh et al[33] resulted in a run time of 2.5 minutes. A short run time of 2.5 minutes can also be obtained using the UPLC-UV assay developed by Padullés et al and UPLC-MS/MS assay developed by Rigo-Bonnin et al.[31,40] The reduction in retention time was aided not only by the intrinsic time of the instruments but also by the implementation of an isocratic program that avoids the time taken to re-equilibrate the chromatographic system.[31,33,40]

Twelve of the 14 summarized studies conducted ganciclovir stability testing in plasma or serum.[25,26,28–34,36,37,40] Plasma samples were stable at room temperature for 16–24 hours,[25,29,30,33,37] whereas serum samples stored

TABLE 1. Characteristics of the Evaluated Assays for the Determination of Ganciclovir and Its Derivatives

Author, Year	Analytes	Instrument	Detection	Sample	Run Time	LLOQ and/or LOD (mg/L)	Stability Testing
Chan et al, 1998[25]	Ganciclovir	HPLC	Spectrofluorimeter (λ_{ex} = 278 nm; λ_{em} = 380 nm)	Serum and heparinized human plasma	≤15 min	0.04	4°C for 0, 24, and 48 h; 22°C for 0, 24, and 48 h
Merodio et al, 2000[26]	Ganciclovir	HPLC	Diode array λ = 254 nm	Albumin nanoparticles; human corneal fibroblasts	8 min	0.05	4°C for 1 mo; −20°C for at least 3 mo
Tsuchie et al, 2001[27]	Ganciclovir	HPLC	Spectrofluorimeter (λ_{ex} = 365 nm; λ_{em} = 512 nm)	Human serum	7.4 min (retention time)	0.005 (LOD)	—
Kishino et al, 2002[28]	Ganciclovir	HPLC	Pulsed amperometer	Plasma samples from transplant recipients	6.26 min	0.01 (LOD), 0.05 (LLOQ)	−20°C for 1 mo
Hosseini et al, 2002[39]	Ganciclovir	Raman spectroscopic system	Raman spectrometer	Rabbit eye	—	—	—
Saleh and Hempel 2006[38]	Ganciclovir	Electrophoresis	UV	Human plasma	—	0.5	—
Perrottet et al, 2007[29]	Ganciclovir	HPLC	Spectrofluorimeter (λ_{ex} = 260 nm; λ_{em} = 380 nm)	Plasma from SOT patients receiving valganciclovir as prophylaxis	13 min (retention time)	0.1	−20°C for at least 4 months; room temperature up to 24 h
Xu et al, 2007[32]	Valganciclovir, ganciclovir	LC-MS/MS	Tandem mass spectrometer	Human plasma	5.5 min	0.004 (valganciclovir), 0.1 (ganciclovir)	3 freeze–thaw cycles; room temperature for 4 h
Weller et al, 2008[30]	Ganciclovir	HPLC	UV	Plasma	8 min	0.05	4°C for 7 d; 23 °C × 24 h; 5 freeze–thaw cycles
Singh et al, 2011[33]	Ganciclovir, valganciclovir	LC-MS/MS	Tandem mass spectrometer	Human plasma	2.5 min	0.005	−20°C and −50°C for 86 d; 3 freeze–thaw cycles stability, bench top stability (25°C) for 16 h, autosampler stability (1–5°C) for 54 h
Padullés et al, 2012[31]	Ganciclovir	UPLC	UV λ = 254 nm	Human plasma	2.5 min	0.5	−20°C for 24 h and 3 mo
Rigo-Bonnin et al, 2014[40]	Ganciclovir	UPLC-MS/MS	Tandem mass spectrometer	Plasma	2.5 min	0.06 (LLOQ), 0.03 (LLOD)	5°C for 7 d; 4°C for 24 h; −75°C for 6 mo
Billat et al, 2015[34]	Ganciclovir and its derivatives in cells	LC-MS/MS	Tandem mass spectrometer	Whole blood healthy volunteers	—	—	At 4°C for 24 h
Gunda et al, 2015[35]	Ganciclovir, valganciclovir, tyrosine-valganciclovir	LC-MS/MS	Tandem mass spectrometer	Rat plasma samples	<3.8 min	0.0005 (ganciclovir), 0.01 (valganciclovir, tyrosine valganciclovir)	—
Märtson et al, 2018[36]	Ganciclovir	LC-MS/MS	Tandem mass spectrometer	Human serum	4.5 min	0.1	20–25°C for 144 h; 4°C for 144 h; 10°C for 120 h; −20°C for 1 yr

TABLE 1. (*Continued*) Characteristics of the Evaluated Assays for the Determination of Ganciclovir and Its Derivatives

Author, Year	Analytes	Instrument	Detection	Sample	Run Time	LLOQ and/or LOD (mg/L)	Stability Testing
Rower et al 2020[37]	Ganciclovir	LC-MS/MS	Tandem mass spectrometer	Dried blood spot from infants	2.4 min (retention time)	0.01	−20°C and −80°C for 1 yr; room temperature for 16 h; autosampler (4°C) for 7 d

HPLC, high-performance liquid chromatography; LLOQ, lower limit of quantification; LOD, lower limit of detection; UPLC, ultra-high–performance liquid chromatography; UV, ultraviolet.

at room temperature were stable for up to 144 hours.[36] In addition, ganciclovir in methanolic stock solution can be stored for up to 1 year at a temperature of −20°C.[36] Ganciclovir in plasma stored at −20°C or −50°C was stable for 86 days.[33] After 5 freeze–thaw cycles, plasma ganciclovir concentrations remained stable.[30]

Rower et al[37] successfully developed and clinically validated an assay to analyze dried blood spots (DBSs), which were extracted using a simple methanol sonication. They confirmed that ganciclovir concentrations in DBSs were similar, correlated well with those observed in serum, and were useful for describing the PK of ganciclovir. Generally, DBS is increasingly being used as an attractive sample for TDM because it offers the benefits of less invasive sample procurement and requirement of low sample volume, which also facilitates transport.[41] Furthermore, it is feasible to perform home sampling with DBS, which would reduce the need for patients to travel to a blood collection site for TDM. The challenges with DBS are impact of spot volume, hematocrit, punch location, and blood collection site on blood volume and uniformity of drug distribution within a DBS punch and clinical validation of the DBS.[42] The assay by Rower et al[37] minimized the effect of spot volume and hematocrit during validation. However, the influence of blood collection sites on ganciclovir assay was not analyzed in this study because all samples were collected using venipuncture.

PRECLINICAL PHARMACOKINETICS AND PHARMACODYNAMICS

Few studies have evaluated the in vitro activity of ganciclovir against CMV. Most models use human embryonic lung or foreskin fibroblast cells, with CMV AD169 as a reference strain. Antiviral activity is expressed in most models as the inhibitory concentration required to reduce viral replication by 50% (IC_{50}). Snoeck et al[43] and Cai et al[44] reported an IC_{50} of 0.6–1.6 mg/L[43] and 0.13–0.2 mg/L,[44] respectively, whereas Freitas et al[45] reported an IC_{50} of 0.9 mg/L (range, 0.6–1).[45] When 42 clinical isolates were evaluated by Balfour et al,[46] a mean IC_{50} of 1.7 μmol/L (range, 0.2–5.3 μmol/L) was observed. When mice were infected with CMV and ganciclovir was evaluated at doses of 1, 3, 9, and 27 mg/kg per day, the median effective dose (ED_{50}, amount of drug that produces a therapeutic response in 50% of the subjects) was 6 mg/kg. In another study, a similar ED_{50} of 7 mg/kg was found in mice.[47]

Similar to other pathogens, the selection of resistant mutants occurs over time when exposed to a drug. The same situation applies to CMV when exposed to ganciclovir.[48] Chou et al[49] tested the IC_{50} of 20 strains with a mutation in the *UL97* gene and found an IC_{50} of >0.7 mg/L (range, 0.8–5.8).[49] Some in vitro studies have also reported the inhibitory concentration required to reduce viral replication by 90% (IC_{90}). Balfour et al[46] reported a mean IC_{90} of 0.3 mg/L (range, 0.1–1).[46] IC_{90} may be a more clinically relevant concentration than the IC_{50} because it better reflects what is aimed for during treatment. Nokta et al[50] showed that 53% inhibition was observed at a concentration of 0.3 mg/L, 74% inhibition at 1.2 mg/L, and 96% inhibition at 3.5 mg/L.[50] Audrey et al used mathematical modeling to analyze the effect of ganciclovir on viral replication.[51] Viral replication was completely inhibited at 20 mg/L, but when balancing efficacy and toxicity, a concentration of 10 mg/L was proposed to be optimal (normalized area under the concentration–time curve (AUC) of viable cells and normalized viral loads for 14 days). Establishing a pharmacokinetic/pharmacodynamic (PK/PD) target for ganciclovir based on preclinical data is difficult because there are no studies combining dynamic drug exposure in relation to inhibition of viral replication. This means that no clues are available on whether the efficacy is concentration-dependent or time-dependent. A potential solution could be to develop a hollow fiber infection model with planktonic fibroblast cells replicating CMV AD169. This could subsequently be exposed to ganciclovir using dose fractionation and could provide the first relevant data for identification of clinical targets.

CLINICAL PHARMACOKINETICS AND PHARMACODYNAMICS

The most common structural population PK model of ganciclovir in both adults and children is a 2-compartment model with lagged first-order absorption (oral administration) and elimination from the central compartment.[10,52–59] A population PK model can be used to calculate the optimal dosing regimen for patients with different characteristics. According to the population PK models, ganciclovir dosing regimens should be based not only on creatinine clearance[52,53,55,57–59] as a surrogate marker for renal function but also on body weight, transplant type, and sex.[56] The population models are listed in Table 2.

The prodrug valganciclovir is rapidly hydrolyzed into ganciclovir after absorption.[60] The time to maximum concentration (t_{max}) of ganciclovir after valganciclovir intake is 1.0–

TABLE 2. Ganciclovir Population Pharmacokinetic Models

Author, Year	Software	Route of Administration or Formulation	Population (n), Country	Final Model
Wiltshire et al, 2005[58]	NONMEM	Oral ganciclovir 1000 mg 3dd and valganciclovir 900 mg 1dd	SOT recipients aged 13 yr and older with a CMV serostatus of D+/R– (n = 364); United Kingdom	CL (L/h) = 12.4 × (CL_{CR}/median)$^{0.925}$ × (WT/79.6)$^{0.725}$ CL_{CR} median males = 80.4 mL/min females = 65.8 mL/min Vc (L) = 25 Vp (L) = 49 Inter-tissue CL (L/h) = 12 tlag (h) = 0.883 Ka (h^{-1}) = 0.128
Chen et al, 2021[53]	NONMEM	Valganciclovir 450 mg and 900 mg 1dd	Adult kidney transplant recipients (n = 70); China	CL (L/h) = 7.09 × (1 + CL_{CR}/68.3 × 1.08) Vc (L) = 10.8 Q (L/h) = 3.96 Vp (L) = 174 Ka (L/h) = 0.23 tlag (h) = 0.93
Czock et al, 2002[54]	WinNonlin	Valganciclovir 900 mg 1dd	HIV-positive and CMV-positive patients (n = 32), healthy volunteers (n = 12); Germany and England	K10 (h^{-1}) = 0.022 K12 (h^{-1}) = 1.44 K21 (h^{-1}) = 0.66 Vc (L/kg) = 0.213 F = 0.63 tlag (h) = 0.77 $tinp_{end}$ (h) = 5.5 Khd (h^{-1}) = 0.57
Zhao et al, 2009[59]	NONMEM	Valganciclovir 900 mg 1dd	Pediatric renal transplant recipients (n = 22); France	CL (L/h) = 8.04 × (CL_{CR}/89)$^{2.93}$ + 3.62 × (WT/28) Vc (L) = 5.2 Vp (L) = 30.7 tlag (h) = 0.743 Ka (h^{-1}) = 0.369
Franck et al, 2020[55]	NONMEM	Valganciclovir 10 mg/kg 2dd and intravenous ganciclovir 5 mg/kg 2dd	Pediatric solid-organ and stem cell transplant recipients (n = 50); Canada	CL × WT/26.7 × CL_{CR}/149.8 (L/h) = 6.9 Vc × WT/26.7 (L) = 9.7 Vp × WT/26.7 (L) = 7.6 Q × WT/26.7 (L) = 10.9 tlag (h) = 0.33 Ka (h^{-1}) = 0.73 F (%) = 43
Vezina et al, 2014[57]	NONMEM	Valganciclovir 900 mg 1dd	Pediatric and adult SOT recipients (n = 82 adults and 13 children); USA	CL/F (L/h) = 14.5 × ((CL_{CR}/60) × (70/WT))$^{0.492}$ × (WT/70)$^{0.75}$ Vc/F (L) = 87.5 × (WT/70) Vp/F (L) = 42.6 × (WT/70) Q/F (L/h) = 4.8 × (WT/70)$^{0.75}$
Perrotet et al, 2009[56]	NONMEM	Valganciclovir 900 mg 2dd (therapy), 900 mg 1dd (prophylaxis), 450 mg 1dd (renal impairment), and intravenous ganciclovir 5 mg/kg 2dd	Adult SOT recipients (n = 65); Switzerland	CL (L/h) = $\theta_{GraftType}$ × GFR_{MDRD} × θ_{female} θ_{kidney} 1.68 θ_{heart} 0.86 $\theta_{lung/liver}$ 1.17 θ_{female} 1.21 Vc (L) = 24 × (WT/70 kg)·θ_{female} θ_{female} 0.78 Vp (L) = 22 Q (L/h) = 4.1 F = 0.6 Ka (h^{-1}) = 0.56

TABLE 2. (*Continued*) Ganciclovir Population Pharmacokinetic Models

Author, Year	Software	Route of Administration or Formulation	Population (n), Country	Final Model
Caldés et al, 2009[52]	NONMEM	Valganciclovir 900 mg 1dd and intravenous ganciclovir 5 mg/kg 2dd	Adult SOT recipients (n = 21); Spain	CL (L/h) = 7.49 × (CL_{CR}/57) Vc (L) = 31.9 Q (L/h) = 10.2 Vp (L) = 32.0 Ka (h^{-1}) = 0.895 F = 0.825 tlag (h) = 0.382
Billat et al, 2016[10]	Pmetrics	Valganciclovir 900 mg 1dd and 450 mg 1dd (renal impairment)	Adult renal transplant recipients (n = 22); France	CL/F (L/h) = 0.58 Vc/F (L) = 32 Vp/F (L) = 40.17 K_{12} (h^{-1}) = 0.016 K_{21} (h^{-1}) = 72.96 tlag = 0.0735

CL, clearance; CL_{CR}, creatinine clearance; WT, body weight; F, bioavailability; Ka, absorption constant; Khd, elimination from the central compartment by hemodialysis; Q, intercompartmental clearance; $tinp_{end}$, time of the end of drug input; tlag, lag time; Vc, central volume of distribution; Vp, peripheral volume of distribution; dd, daily dose.

3.5 hours.[54,61–65] The bioavailability of valganciclovir is 24%–56% higher in the fed condition than that in the fasted condition.[66] In addition, food delays the t_{max} of ganciclovir after valganciclovir intake, especially at higher dosages, with the respective fasted and fed t_{max} being 1–1.8 hours and 1.5–2 hours, respectively.[66] Plasma protein binding of ganciclovir is negligible (1%–2%) over the concentration range of 0.5–51 mg/L.[9]

Ganciclovir is eliminated mainly through the kidneys by glomerular filtration and active tubular secretion. In patients with normal renal function, i.v. ganciclovir is 90% unchanged when it is excreted in the urine.[9] Elimination of ganciclovir is biphasic, and both systemic and intercompartmental clearances have been estimated in various studies.[10,52–59,67] In patients with mild renal impairment, ganciclovir clearance is almost half of the clearance value in healthy subjects (CL/F 14.9 L/h *vs.* 24.2 L/h).[54] Similarly, the mean ganciclovir clearance in renal transplant patients is lower (CL 0.6 L/h)[10] than that in other transplant recipients (CL mean 11.15 L/h).[10,52,53,55,57–59,68,69]

Apart from renal function, other factors may also affect ganciclovir elimination. Differences in drug regimens for specific transplantations (eg, immunosuppressives) may contribute to the variability in ganciclovir elimination.[56] Interestingly, female patients have a higher clearance than men, which may be associated with the sex differences in organic anion transporter expression observed in rodents.[56,70,71]

Currently, there are limited data available regarding the exposure targets to use to optimize therapy. In addition, the specific IC_{90} that is related to the decrease in viral load in patients has not been confirmed. A target 24-hour area under the time concentration curve (AUC_{24h}) of 40–60 mg·h/L has been proposed for prophylaxis.[72,73] Wiltshire et al described that an AUC of 40–50 mg·h/L was associated with a suppression of viral load during prophylaxis after 1 month; however, this was not seen after 6 months and they did not evaluate other PK parameters besides the AUC.[73] Stockmann et al suggested in an expert opinion that an AUC_{24h} of 80–120 mg·h/L could be used as a potential efficacy target to treat CMV infections.[72] Although these AUC_{24h} targets have been used to optimize therapy, no PK/PD index is available to improve efficacy and reduce toxicity.[74]

THERAPEUTIC DRUG MONITORING

There is an urgent need for optimization of ganciclovir dosing to avoid antiviral resistance and toxicity, especially in HSCT recipients.[17,72,75] Various case studies have presented TDM as a potential solution to optimize treatment in specific clinical scenarios.[76–78] Despite the lack of strong evidence to support TDM, multiple centers have started TDM programs.[52,74,79–81] In these studies, specific target ranges were defined. Richie et al used 1–3 mg/L for C_{min} and 3–12.5 mg/L for C_{max}, whereas Märtson et al defined $AUC_{24h} > 50$ mg·h/L or C_{min} of 1–2 mg/L for prophylaxis and 80–120 mg·h/L or 2–4 mg/L for treatment, respectively.[74,79] These targets were based on either expert opinions or calculations from the IC_{50} of ganciclovir.

A retrospective study on ganciclovir TDM by Ritchie et al[79] reported 82 patients with CMV infection and observed large interindividual variability among them.[79] Moreover, 52% of these patients did not reach the predefined target ranges.[79] No relationship was found between drug exposure and treatment efficacy or toxicity.[79] Similarly, high interindividual and intraindividual variability was observed in a study performed in 95 transplant recipients, where patients on both prophylaxis and treatment of CMV and herpesvirus type 6 were included.[74] It was also seen that even appropriate dosing results in underexposure in both the prophylaxis and treatment groups and that the AUC did not have a strong correlation with C_{min} values.[74] This could mean that C_{min} alone does not provide a good overview of ganciclovir exposure. In addition, underexposure was observed in patients with an estimated glomerular filtration rate (eGFR) > 90 mL/min/1.73 m^2, which could be expected because of the PK of ganciclovir.[74] The decrease in white blood cell counts significantly correlated with the highest AUC and C_{min} values, which could mean that toxicity could

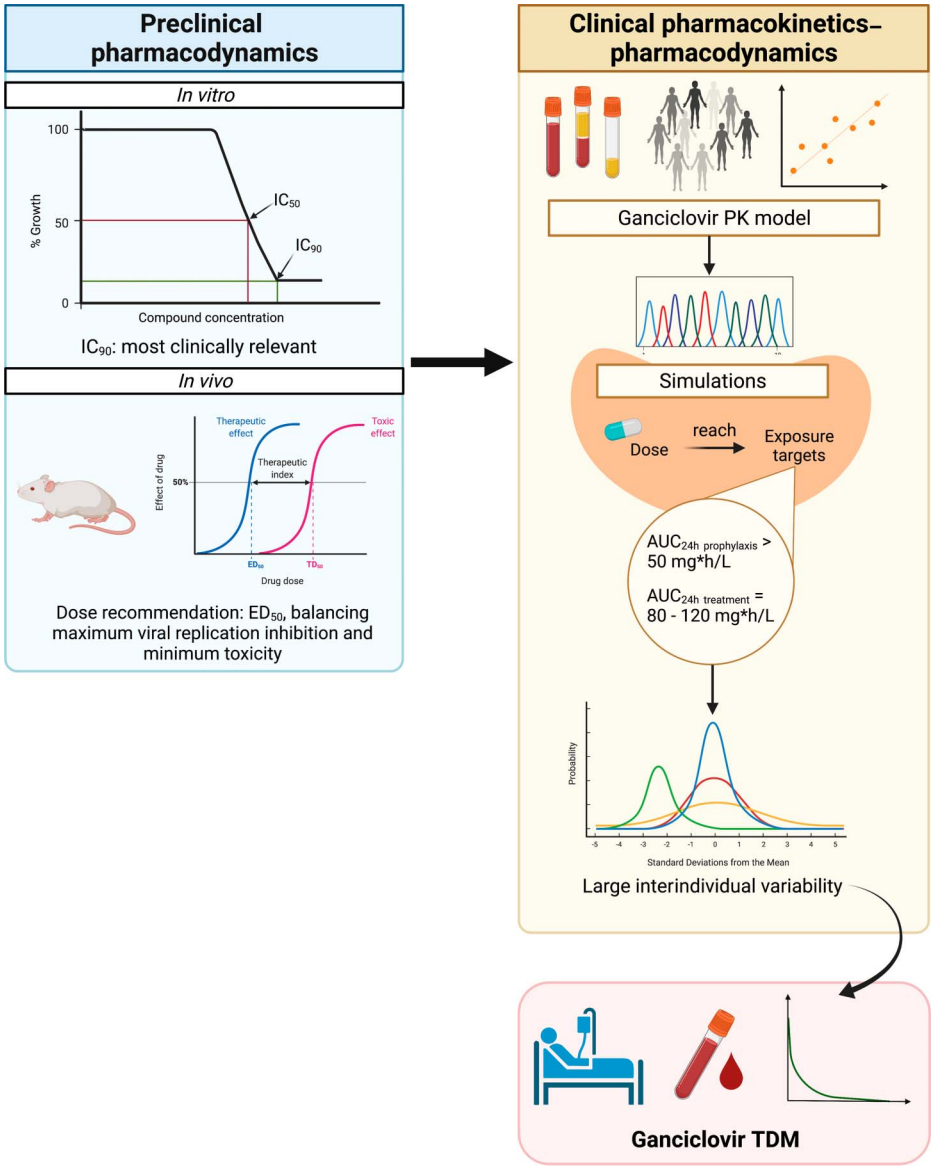

FIGURE 2. Preclinical and clinical PK/PD and TDM of ganciclovir.

be suspected with higher concentrations and AUC values. Because of the observational nature of the study and lack of follow-up data, clinical outcomes could not be linked to ganciclovir exposure. In another recent study, 90 patients with CMV infection were evaluated during a 17-month study period.[82] Although patients did not always receive a dose according to guideline, the study showed that with peak concentrations lower than 8.37 mg/L or higher than 11.86 mg/L, poorer outcomes were observed.[82] Poor outcomes were defined as time to resolution of CMV, breakthrough CMV during prophylaxis, and cessation of therapy due to toxicity.[82] The preclinical and clinical PK and PD, together with potential TDM applications, are shown in Figure 2.

In the pediatric population, underexposure and variability of concentrations have also been observed, and the application of the same AUC_{24h} targets has been used; however, there is a need for more studies on children.[72,80,83] Åsberg et al proposed a new ganciclovir dosing algorithm in pediatric SOT recipients using nonparametric modeling, where they conducted Monte Carlo simulations to evaluate the new regimens.[83] The algorithm included body weight and not body surface area. In addition, the renal function (eGFR) was estimated using the Cockcroft–Gault formula as opposed to the regularly used Schwartz formula in children.[83] The model accurately predicted ganciclovir concentrations.

DISCUSSION, GAP ANALYSIS, AND OUTLOOK

(Val)ganciclovir therapy is complicated by frequently reported toxicities. The data on ganciclovir concentrations show large interindividual variability, indicating that TDM may play a role in modifying the dose to reduce toxicity and prevent treatment failure related to low concentrations. The main hurdle for implementing TDM is the lack of robust data to define a therapeutic window. Before TDM-guided

intervention studies can be performed, there is a need to better understand the relationship between ganciclovir exposure and the inhibition of viral replication by intracellular ganciclovir triphosphate.[75,84] In addition to the PK/PD of ganciclovir, there is a need for a better understanding of the quantitative role of the immune response of the patient and exposure to different immunosuppressives in relation to viral clearance.[75,81,85] Ho et al[75] formulated that the interplay between PK, PD, and host immunity factors should be the focus of future research. Indeed, it is important to consider all these when designing studies for optimization of ganciclovir therapy because the studies conducted to date have mainly focused on estimating ganciclovir exposure, and owing to the limited therapeutic options for CMV infections, future studies on ganciclovir are warranted.

ACKNOWLEDGMENTS

Figures 1 and 2 were created with Biorender.com.

REFERENCES

1. Kotton CN. CMV: prevention, diagnosis and therapy. *Am J Transpl*. 2013;13(suppl 3):24–40.
2. Kotton CN, Kumar D, Caliendo AM, et al. The third international consensus guidelines on the management of cytomegalovirus in solid-organ transplantation. *Transplantation*. 2018;102:900–931.
3. Pellett Madan R, Hand J. Human herpesvirus 6, 7, and 8 in solid organ transplantation: guidelines from the american society of transplantation infectious diseases community of practice. *Clin Transpl*. 2019;33:e13518.
4. Beigel JH. Antiviral therapy (Non-HIV). *Goldman's Cecil Med*. 2012; 2012:2082–2089.
5. Roche. Valcyte 450 mg film-coated tablets, SmPC. Available at: https://www.medicines.org.uk/emc/product/1641/smpc. Accessed January 9, 2021.
6. Biron KK, Stanat SC, Sorrell JB, et al. Metabolic activation of the nucleoside analog 9-[(2-hydroxy-1-(hydroxymethyl)ethoxy]methyl)guanine in human diploid fibroblasts infected with human cytomegalovirus. *Proc Natl Acad Sci U S A*. 1985;82:2473–2477.
7. Razonable RR. Antiviral drugs for viruses other than human immunodeficiency virus. *Mayo Clinic Proceedings*. 2011;86:1009–1026.
8. Sullivan V, Talarico CL, Stanat SC, et al. A protein kinase homologue controls phosphorylation of ganciclovir in human cytomegalovirus-infected cells. *Nature*. 1992;359:162–164.
9. FDA. Ganciclovir summary of product characteristics. Available at: www.fda.gov/medwatch.
10. Billat PA, Woillard JB, Essig M, et al. Plasma and intracellular exposure to ganciclovir in adult renal transplant recipients: is there an association with haematological toxicity? *J Antimicrob Chemother*. 2016;71:484–489.
11. Razonable RR, Hayden RT. Clinical utility of viral load in management of cytomegalovirus infection after solid organ transplantation. *Clin Microbiol Rev*. 2013;26:703–727.
12. Ljungman P, Griffiths P, Paya C. Definitions of cytomegalovirus infection and disease in transplant recipients. *Clin Infect Dis*. 2002;34:1094–1097.
13. Razonable RR, Humar A. Cytomegalovirus in solid organ transplant recipients—guidelines of the american society of transplantation infectious diseases community of practice. *Clin Transpl*. 2019;33:e13512.
14. Humar A, Lebranchu Y, Vincenti F, et al. The efficacy and safety of 200 days valganciclovir cytomegalovirus prophylaxis in high-risk kidney transplant recipients. *Am J Transpl*. 2010;10:1228–1237.
15. Takahata M, Hashino S, Nishio M, et al. Occurrence of adverse events caused by valganciclovir as pre-emptive therapy for cytomegalovirus infection after allogeneic stem cell transplantation is reduced by low-dose administration. *Transpl Infect Dis*. 2015;17:810–815.
16. Paya C, Humar A, Dominguez E, et al. Efficacy and safety of valganciclovir vs. oral ganciclovir for prevention of cytomegalovirus disease in solid organ transplant recipients. *Am J Transpl*. 2004;4:611–620.
17. Venton G, Crocchiolo R, Fürst S, et al. Risk factors of ganciclovir-related neutropenia after allogeneic stem cell transplantation: a retrospective monocentre study on 547 patients. *Clin Microbiol Infect*. 2014;20:160–166.
18. McGavin JK, Goa KL. Ganciclovir: an update of its use in the prevention of cytomegalovirus infection and disease in transplant recipients. *Drugs*. 2001;61:1153–1183.
19. Faulds D, Heel RC. Ganciclovir. A review of its antiviral activity, pharmacokinetic properties and therapeutic efficacy in cytomegalovirus infections. *Drugs*. 1990;39:597–638.
20. Turgeon N, Hovingh GK, Fishman JA, et al. Safety and efficacy of granulocyte colony-stimulating factor in kidney and liver transplant recipients. *Transpl Infect Dis*. 2000;2:15–21.
21. Biron KK. Antiviral drugs for cytomegalovirus diseases. *Antivir Res*. 2006;71:154–163.
22. Gilbert C, Boivin G. Human cytomegalovirus resistance to antiviral drugs. *Antimicrob Agents Chemother*. 2005;49:873–883.
23. Lurain NS, Chou S. Antiviral drug resistance of human cytomegalovirus. *Clin Microbiol Rev*. 2010;23:689–712.
24. Gagermeier JP, Rusinak JD, Lurain NS, et al. Subtherapeutic ganciclovir (GCV) levels and GCV-resistant cytomegalovirus in lung transplant recipients. *Transpl Infect Dis*. 2014;16:941–950.
25. Chan R, Kiang CH, Chu F, et al. A rapid, sensitive HPLC method for the determination of ganciclovir in human plasma and serum. *J Pharm Belg*. 1998;53:170.
26. Merodio M, Campanero MA, Mirshahi T, et al. Development of a sensitive method for the determination of ganciclovir by reversed-phase high-performance liquid chromatography. *J Chromatogr A*. 2000;870: 159–167.
27. Tsuchie M, Hara S, Kimura M, et al. Determination of anti-virus drug, ganciclovir, in human serum by HPLC with precolumn fluorescence derivatization using phenylglyoxal. *Anal Sci*. 2001;17:811–814.
28. Kishino S, Takekuma Y, Sugawara M, et al. Liquid chromatographic method for the determination of ganciclovir and/or acyclovir in human plasma using pulsed amperometric detection. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2002;780:289–294.
29. Perrottet N, Beguin A, Meylan P, et al. Determination of aciclovir and ganciclovir in human plasma by liquid chromatography-spectrofluorimetric detection and stability studies in blood samples. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2007;852:420–429.
30. Weller DR, Balfour HH, Vezina HE. Simultaneous determination of acyclovir, ganciclovir, and (R)-9-[4-hydroxy-2-(hydroxymethyl)butyl] guanine in human plasma using high-performance liquid chromatography. *Biomed Chromatogr*. 2009;23:822–827.
31. Padullés A, Colom H, Armendariz Y, et al. Determination of ganciclovir in human plasma by ultra performance liquid chromatography–UV detection. *Clin Biochem.*. 2012;45:309–314.
32. Xu HR, Li XN, Chen WL, et al. A sensitive assay for simultaneous determination of plasma concentrations of valganciclovir and its active metabolite ganciclovir by LC/MS/MS. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2007;848:329–334.
33. Singh O, Saxena S, Mishra S, et al. Determination of valganciclovir and ganciclovir in human plasma by liquid chromatography tandem mass spectrometric detection. *Clin Biochem*. 2011;44:907–915.
34. Billat P, Sauvage F, Picard N, et al. Liquid chromatography tandem mass spectrometry quantitation of intracellular concentrations of ganciclovir and its phosphorylated forms. *Fundam Clin Pharmacol*. 2014;28:30–90.
35. Gunda S, Earla R, Cholkar K, et al. Pharmacokinetic studies and LC-MS/MS method development of ganciclovir and dipeptide monoester prodrugs in Sprague Dawley rats. *Eur J Drug Metab Pharmacokinet*. 2015; 40:325–334.
36. Märtson A, van Hateren K, van den Bosch G, et al. Determination of ganciclovir and acyclovir in human serum using liquid chromatography-tandem mass spectrometry. *J Appl Bioanalj Appl Bio*. 2018:175–186.
37. Rower JE, Nielson C, Shi K, et al. Validation of an assay for quantifying ganciclovir in dried blood spots. *J Pharm Biomed Anal*. 2020;184: 113181.
38. Saleh S, Hempel G. Quantification of ganciclovir in human plasma using capillary electrophoresis. *Electrophoresis*. 2006;27:2439–2443.

39. Hosseini K, March W, Jongsma FH, et al. Noninvasive detection of ganciclovir in ocular tissue by Raman spectroscopy: implication for monitoring of drug release. *J Ocul Pharmacol Ther*. 2002;18:277–285.
40. Rigo-Bonnin R, Padullés A, Corral-Comesaña S, et al. Measurement of ganciclovir concentration in human plasma by ultra-performance liquid chromatography–tandem mass spectrometry. *Clin Chim Acta*. 2014;427:58–64.
41. De Kesel PMM, Sadones N, Capiau S, et al. Hemato-critical issues in quantitative analysis of dried blood spots: challenges and solutions. *Bioanalysis*. 2013;5:2023–2041.
42. Capiau S, Veenhof H, Koster RA, et al. Official international association for therapeutic drug monitoring and clinical toxicology guideline: development and validation of dried blood spot-based methods for therapeutic drug monitoring. *Ther Drug Monit*. 2019;41:409–430.
43. Snoeck R, Andrei G, Schols D, et al. Activity of different antiviral drug combinations against human cytomegalovirus replication in vitro. *Eur J Clin Microbiol Infect Dis*. 1992;11:1144–1155.
44. Cai H, Kapoor A, He R, et al. In vitro combination of anti-cytomegalovirus compounds acting through different targets: role of the slope parameter and insights into mechanisms of action. *Antimicrob Agents Chemother*. 2014;58:986–994.
45. Freitas VR, Fraser-Smith EB, Chiu S, et al. Efficacy of ganciclovir in combination with zidovudine against cytomegalovirus in vitro and in vivo. *Antivir Res*. 1993;21:301–315.
46. Balfour HH, Boivin G, Erice A, et al. Ganciclovir susceptibilities of cytomegalovirus (CMV) isolates from solid organ transplant recipients with CMV viremia after antiviral prophylaxis. *J Infect Dis.*. 1993;168:332–335.
47. Freitas VR, Fraser-Smith EB, Matthews TR. Efficacy of ganciclovir in combination with other antimicrobial agents against cytomegalovirus in vitro and in vivo. *Antivir Res*. 1993;20:1–12.
48. Drew WL, Paya CV, Emery V. Cytomegalovirus (CMV) resistance to antivirals. *Am J Transpl*. 2001;1:307–312.
49. Chou S, Guentzel S, Michels KR, et al. Frequency of UL97 phosphotransferase mutations related to ganciclovir resistance in clinical cytomegalovirus isolates. *J Infect Dis*. 1995;172:239–242.
50. Nokta M, Tolpin MD, Nadler PI, et al. Human monoclonal anti-cytomegalovirus (CMV) antibody (MSL 109): enhancement of in vitro foscarnet- and ganciclovir-induced inhibition of CMV replication. *Antivir Res*. 1994;24:17–26.
51. Janoly-Dumenil A, Rouvet I, Bleyzac N, et al. A pharmacodynamic model of ganciclovir antiviral effect and toxicity for lymphoblastoid cells suggests a new dosing regimen to treat cytomegalovirus infection. *Antimicrob Agents Chemother*. 2012;56:3732–3738.
52. Caldés A, Colom H, Armendariz Y, et al. Population pharmacokinetics of ganciclovir after intravenous ganciclovir and oral valganciclovir administration in solid organ transplant patients infected with cytomegalovirus. *Antimicrob Agents Chemother*. 2009;53:4816–4824.
53. Chen B, Hu SS, Rui WB, et al. Population pharmacokinetics and bayesian estimation of the area under the concentration-time curve for ganciclovir in adult Chinese renal allograft recipients after valganciclovir administration. *J Clin Pharmacol*. 2021;61:328–338.
54. Czock D, Scholle C, Rasche FM, et al. Pharmacokinetics of valganciclovir and ganciclovir in renal impairment. *Clin Pharmacol Ther*. 2002;72:142–150.
55. Franck B, Woillard JB, Théorêt Y, et al. Population pharmacokinetics of ganciclovir and valganciclovir in paediatric solid organ and stem cell transplant recipients. *Br J Clin Pharmacol*. 2021;87:3105–3114.
56. Perrottet N, Csajka C, Pascual M, et al. Population pharmacokinetics of ganciclovir in solid-organ transplant recipients receiving oral valganciclovir. *Antimicrob Agents Chemother*. 2009;53:3017–3023.
57. Vezina HE, Brundage RC, Balfour HH. Population pharmacokinetics of valganciclovir prophylaxis in paediatric and adult solid organ transplant recipients. *Br J Clin Pharmacol*. 2014;78:343–352.
58. Wiltshire H, Hirankarn S, Farrell C, et al. Pharmacokinetic profile of ganciclovir after its oral administration and from its prodrug, valganciclovir, in solid organ transplant recipients. *Clin Pharmacokinet*. 2005;44:495–507.
59. Zhao W, Baudouin V, Zhang D, et al. Population pharmacokinetics of ganciclovir following administration of valganciclovir in paediatric renal transplant patients. *Clin Pharmacokinet.*. 2009;48:321–328.
60. Cvetkovi RS, Wellington K. Valganciclovir: a review of its use in the management of CMV infection and disease in immunocompromised patients. *Drugs*. 2005;65:859–878.
61. Jung D, Dorr A. Single-dose pharmacokinetics of valganciclovir in HIV- and CMV-seropositive subjects. *J Clin Pharmacol*. 1999;39:800–804.
62. Kimberlin DW, Acosta EP, Sánchez PJ, et al. Pharmacokinetic and pharmacodynamic assessment of oral valganciclovir in the treatment of symptomatic congenital cytomegalovirus disease. *J Infect Dis*. 2008;197:836–845.
63. Kiser TH, Fish DN, Zamora MR. Evaluation of valganciclovir pharmacokinetics in lung transplant recipients. *J Heart Lung Transpl*. 2012;31:159–166.
64. 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.
65. Winston DJ, Baden LR, Gabriel DA, et al. Pharmacokinetics of ganciclovir after oral valganciclovir versus intravenous ganciclovir in allogeneic stem cell transplant patients with graft-versus-host disease of the gastrointestinal tract. *Biol Blood Marrow Transpl*. 2006;12:635–640.
66. Brown F, Banken L, Saywell K, et al. Pharmacokinetics of valganciclovir and ganciclovir following multiple oral dosages of valganciclovir in HIV- and CMV-seropositive volunteers. *Clin Pharmacokinet*. 1999;37:167–176.
67. Horvatits T, Kitzberger R, Drolz A, et al. Pharmacokinetics of ganciclovir during continuous venovenous hemodiafiltration in critically ill patients. *Antimicrob Agents Chemother*. 2014;58:94–101.
68. Jung D, Abdel-Hameed MH, Teitelbaum P, et al. The pharmacokinetics and safety profile of oral ganciclovir in combination with zalcitabine or stavudine in asymptomatic HIV- and CMV-seropositive patients. *J Clin Pharmacol*. 1999;39:505–512.
69. Cimoch P, Lavelle J, Pollard R, et al. Pharmacokinetics of oral ganciclovir alone in combination with zidovudine, didanosine and probenecid in HIV-infected subjects. *J Acquir Immune Defic Syndr Hum Retrovirology*. 1998;17:227–234.
70. Ljubojević M, Herak-Kramberger CM, Hagos Y, et al. Rat renal cortical OAT1 and OAT3 exhibit gender differences determined by both androgen stimulation and estrogen inhibition. *Am J Physiol Ren Physiol.*. 2004;287:F124–F138.
71. Groves CE, Suhre WB, Cherrington NJ, et al. Sex differences in the mRNA, protein, and functional expression of organic anion transporter (Oat) 1, Oat3, and organic cation transporter (Oct) 2 in rabbit renal proximal tubules. *J Pharmacol Exp Ther*. 2006;316:743–752.
72. Stockmann C, Roberts JK, Knackstedt ED, et al. Clinical pharmacokinetics and pharmacodynamics of ganciclovir and valganciclovir in children with cytomegalovirus infection. *Expert Opin Drug Metab Toxicol*. 2015;11:205–219.
73. Wiltshire H, Paya CV, Pescovitz MD, et al. Pharmacodynamics of oral ganciclovir and valganciclovir in solid organ transplant recipients. *Transplantation*. 2005;79:1477–1483.
74. Märtson AG, Edwina AE, Burgerhof JGM, et al. Ganciclovir therapeutic drug monitoring in transplant recipients. *J Antimicrob Chemother*. 2021;76:2356–2363.
75. Ho SA, Slavin M, Roberts JA, et al. Optimization of Ganciclovir use in allogeneic hematopoietic cell transplant recipients—the role of therapeutic drug monitoring. *Expert Rev Anti Infect Ther*. 2020;19:707–718.
76. Märtson AG, Touw D, Damman K, et al. Ganciclovir therapeutic drug monitoring: a case series. *Ther Drug Monit*. 2019;41:107–110.
77. Peredo I, Hellden A, Wolmer-Solberg N, et al. Ganciclovir concentrations in the cerebral extracellular space after valganciclovir treatment; a case study. *BMJ Case Rep*. 2015;2015:1–4.
78. Nunez-Nunez M, Bellapart J, O'Donoghue S, et al. Variable ganciclovir concentrations in a critically ill patient receiving continuous renal replacement therapy and plasma exchange? *Int J Antimicrob Agents*. 2014;43:572–573.
79. Ritchie BM, Barreto JN, Barreto EF, et al. Relationship of ganciclovir therapeutic drug monitoring with clinical efficacy and patient safety. *Antimicrob Agents Chemother*. 2019;63:e01855–18.
80. Luck S, Lovering A, Griffiths P, et al. Ganciclovir treatment in children: evidence of subtherapeutic levels. *Int J Antimicrob Agents*. 2011;37:445–448.
81. Perrottet N, Manuel O, Lamoth F, et al. Variable viral clearance despite adequate ganciclovir plasma levels during valganciclovir treatment for

cytomegalovirus disease in D+/R- transplant recipients. *BMC Infect Dis*. 2010;10:2.
82. Galar A, Valerio M, Catalán P, et al. Valganciclovir-ganciclovir use and systematic therapeutic drug monitoring. An invitation to antiviral stewardship. *Antibiotics (Basel)*. 2021;10:77.
83. Åsberg A, Bjerre A, Neely M. New algorithm for valganciclovir dosing in pediatric solid organ transplant recipients. *Pediatr Transpl*. 2014;18:103–111.
84. Ljungman P, Boeckh M, Hirsch HH, et al. Definitions of cytomegalovirus infection and disease in transplant patients for use in clinical trials. *Clin Infect Dis*. 2017;64:87–91.
85. San Juan R, Aguado JM, Lumbreras C, et al. Impact of current transplantation management on the development of cytomegalovirus disease after renal transplantation. *Clin Infect Dis*. 2008;47:875–882.