SUPPLEMENTAL MATERIAL

METHODS

CMV disease definitions. For the purposes of this study, CMV syndrome was defined as the presence of CMV in the blood and at least one of the following: fever, new or increased malaise, leukopenia, atypical lymphocytosis, or thrombocytopenia. Tissue-invasive CMV disease was defined as presence of CMV in the blood and at least one of the following: localized CMV infection confirmed in a biopsy or other specimen and relevant symptoms or signs of organ dysfunction unlikely to be due to other causes (S1).

GCV resistance testing. Plasma samples were sent for resistance testing in patients who 1) initiated antiviral therapy and did not demonstrate a reduction in CMV viral load after 2 weeks on therapy; 2) experienced virologic rebound defined as a) confirmed (defined as ≥2 viral load readings) increase in viremia of ≥1 $\log_{10}$ from nadir while on therapy or b) confirmed detectable (≥150 copies/mL; 137 IU/mL) CMV viral load on therapy after confirmed undetectable CMV viral load; 3) had a confirmed diagnosis of CMV disease with a detectable (≥150 copies/mL; 137 IU/mL) viral load; or 4) had a CMV viral load >1,000 copies/mL (910 IU/mL) after antiviral treatment completed at any time point. Viral UL97 kinase and UL54 DNA polymerase gene mutations are known to confer resistance to ganciclovir (S2), and ganciclovir resistance testing was performed by sequencing regions of the UL97 and UL54 genes containing known variants (Viracor-IBT Laboratories, Lee's Summit, MO).

Pharmacokinetics and immunogenicity assessments. Serum samples for pharmacokinetic analysis were collected up to 24 hours prior to first dosing (day 1); 1 hour (± 15 min), 4 hours (± 2 hours), 24 hours (day 2) and 72 hours (day 4) after first dosing; on day 8 before dosing and 1 hour (± 15 min) after dosing; on day 29 before dosing and 1 hour (± 15 min) after dosing; on day 57 before dosing and 1 hour (± 15 min) and 24 hours (day 58) after dosing; on days 43, 64, 71, 78, 85, 113 and 141; and at study completion (day 169)/early termination. Serum samples for the detection of anti-therapeutic antibodies (ATAs) were collected on days 1 (pre-dose), 29 (pre-dose), 57 (pre-dose), 85, 113, 141 and at study completion (day 169)/early termination.

Sample size calculation/statistical analysis. Assuming that 55% of the RG7667 group and 75%–80% of the placebo group would have CMV viremia within the first 12 weeks posttransplant (S3-S5), a sample size of 50 patients per group was estimated to provide 90% confidence interval (CI) widths of ±12% and ±10% for CMV viremia in the RG7667 and control groups, respectively, and a 95% CI for the difference between groups of ±18%. In order to account for a 10% dropout rate for the primary endpoint and an estimated 10% of patients who may be CMV seropositive (D+R+) at baseline because of seroconversion since the initial serological evaluation, 60 patients per group were estimated to be required. The emphasis of this study was on effect size estimation rather than formal hypothesis testing, so no adjustment for a type 1 error was made to account for the multiplicity of analyses.

Patients who withdrew prior to week 12 or 24 with a positive CMV viral load <150 copies/mL (137 IU/mL) were considered to have a detectable viral load for the week 12 and 24 CMV viremia efficacy analyses, respectively. Pre-specified subgroup analyses according to region (United States versus European Union) and use of anti-thymocyte globulin and/or alemtuzumab for induction immunosuppression were performed for the week 12 CMV viremia efficacy analysis to examine the consistency of the treatment estimates. For time-to-event analyses, data for patients without detectable CMV viremia were censored at the date of the last viral load assessment. Viral load data were $\log_{10}$ transformed prior to analysis.

RESULTS

Pharmacokinetics and immunogenicity results. Serum MCMV5322A and MCMV3068A concentrations exhibited a biphasic disposition, with an initial rapid distribution phase followed by a slow elimination phase (Fig S1). MCMV5322A and MCMV3068A exposure was confirmed in all RG7667-treated patients. The mean terminal half-lives for MCMV5322A and MCMV3068A in our population of kidney transplant recipients were 26.9 and 27.4 days, respectively, which were consistent with the half-lives observed in healthy subjects (S6). Patients with evaluable immunogenicity data (i.e., with at least one post-baseline sample) were considered to have developed an ATA response during the study if they tested negative at baseline and positive after dosing. Eight of 56 (14.3%) RG7667-treated patients with evaluable immunogenicity data developed an ATA response to only MCMV5322A (n = 3), only MCMV3068A (n = 4) or both antibodies (n = 1). Four of 59 (6.8%) placebo-treated patients with evaluable immunogenicity data developed an ATA response.

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TABLE S1 Proportion with CMV viremia[a] within 12 weeks after transplant by region and induction immunosuppression

	Number of Patients with CMV Viremia/ Number of Patients within Strata (%)	
	RG7667 (n = 59)	Placebo (n = 57)
Overall	27/59 (45.8)	35/57 (61.4)
Region		
United States	12/28 (42.8)	15/27 (55.6)
European Union	15/31 (48.3)	20/30 (66.7)
ATG[b] and/or alemtuzumab for induction immunosuppression		
Yes	6/14 (42.9)	12/15 (80.0)
No	21/45 (46.7)	23/42 (54.8)

[a]Viral load ≥150 copies/mL (137 IU/mL).
[b]ATG, anti-thymocyte globulin.

TABLE S2 Median time to CMV viremia[a] by region and induction immunosuppression

	Days	
	RG7667 (n = 59)	Placebo (n = 57)
Overall	139	46
Region		
United States	139	70
European Union	83	42
ATG[b] and/or alemtuzumab for induction immunosuppression		
Yes	58	42
No	160	49

[a]Viral load ≥150 copies/mL (137 IU/mL).
[b]ATG, anti-thymocyte globulin.

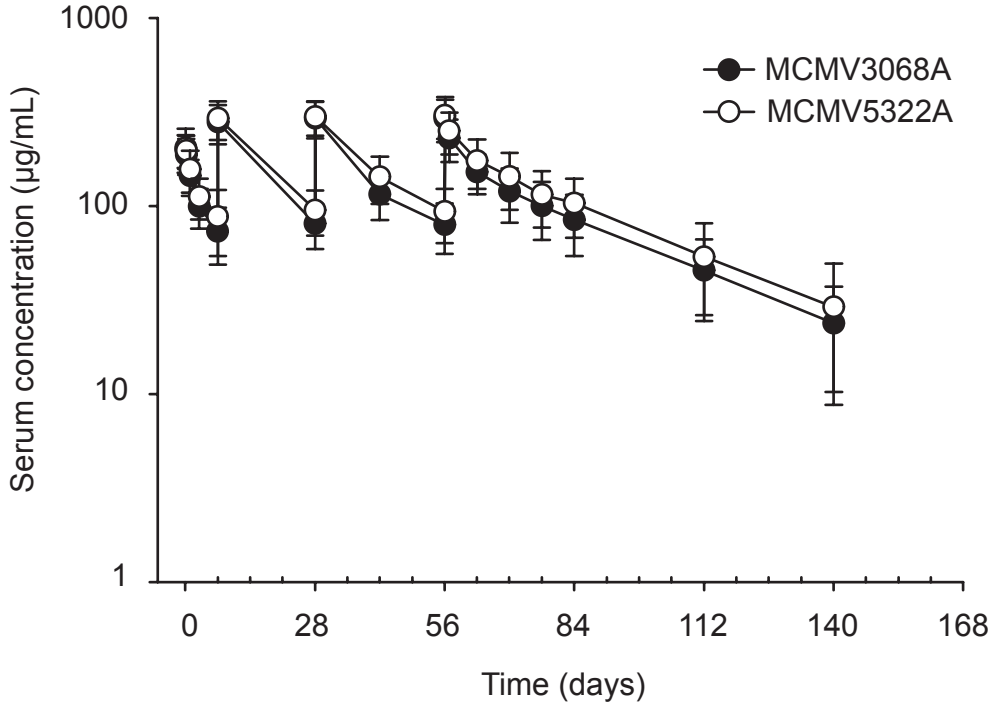

FIG S1 Group mean (± SD) MCMV5322A and MCV3068A serum concentration versus time profiles following intravenous administration of RG7667 (10 mg/kg of each antibody) on Day 1, 8, 29, and 57.

Note: Nominal sampling time was used for plotting.