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ORIGINAL ARTICLE

Molecular characterization of cytomegalovirus based on glycoprotein B and N among solid organ and hematopoietic stem cell transplant recipients in Jordan

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ABSTRACT

Background and Objectives: Cytomegalovirus (CMV), a prevalent member of the herpesvirus family, poses significant risks to immunocompromised patients, particularly those undergoing hematopoietic stem cell transplantation (HSCT) or solid organ transplantation (SOT). This study aimed to assess the prevalence and genotype distribution of CMV among transplant recipients in Jordan.

Materials and Methods: A retrospective observational study conducted at the Jordan Royal Medical Service's Virology Department from January to October 2024, included all patients who underwent HSCT or SOT. Blood samples collected in EDTA tubes were analyzed for CMV detection and genotyping. Real-time PCR facilitated CMV amplification, while multiplex nested PCR identified gB and gN genotypes.

Results: Among 80 transplant recipients with positive CMV DNA, 15 (18.8%) were from SOT kidney transplants (KT), and 65 (81.2%) were HSCT recipients. Genotype analysis of 44 samples revealed that 21 had the gN genotype and 27 had the gB genotype. Mixed genotypes gB and gN were present in 15 samples. The mixed genotype gN1+gN2 (42.86%) was most common in KT recipients, while gB2 (31%) was prevalent among HSCT recipients.

Conclusion: CMV is a common opportunistic virus that often leads to severe, life-threatening illness and is associated with an increased risk of transplant rejection. Our study demonstrated that the most prevalent genotypes in Jordanian HSCT and SCT recipients with CMV infection were gB2 and gN1+gN2, respectively.

Keywords: Cytomegalovirus; Glycoproteins; Glycoprotein B; Glycoprotein N; Solid organ transplantation; Hematopoietic stem cell transplantation; Immunocompromised host

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INTRODUCTION

Cytomegalovirus (CMV) is a member of the *Herpesviridae* family and the *Betaherpesvirinae* subfamily (1). It is notable for being the largest DNA virus among herpes viruses, measuring approximately 150-200 nm in size and containing a linear double-stranded DNA genome within its nucleocapsid (2). CMV can infect individuals of all ages, leading to a range of symptoms and complications, including low birth weight, hepatosplenomegaly, and various developmental defects affecting mental, cardiovascular, and muscular systems (3). The estimated global seroprevalence of CMV in the general population is around 83%, though this figure varies by region, with the Eastern Mediterranean region having the highest prevalence at approximately 90% and Europe showing a lower rate of around 66% (4).

One reason for CMV's widespread infection is its broad cell tropism, enabling it to spread easily within the host. The virus can replicate in numerous cell types, including fibroblasts, smooth muscle cells, endothelial cells, connective tissue, epithelial tissue, and hematopoietic stem cells (5). Infection can manifest as primary infection, reinfection, or reactivation, posing significant risks, particularly to transplant recipients, immunodeficient patients, and those undergoing immunosuppressive treatment (6). Following an initial infection, CMV establishes a lifelong latent infection that may reactivate or lead to reinfection with different strains (7).

The variability observed among CMV strains is largely attributed to its glycoproteins, including glycoprotein B (gB), gM, gN, gH, gL, and gO, which are encoded by distinct genes (8). These glycoproteins play crucial roles in the virus's pathophysiology, facilitating cell penetration, cell-to-cell transmission, immune response activation, and the expression of gB on the viral surface, which serves as a target for neutralizing antibodies and provides protective effects (9, 10). Specifically, the CMV UL55 gene encodes gB, while the UL73 gene encodes gN. Variability in the UL55 gene reveals four primary genotypes (gB1, gB2, gB3, and gB4), in addition to three non-prototypic genotypes (gB5, gB6, and gB7) (8). Similarly, polymorphisms in the UL73 gene indicate four main genotypes (gN1, gN2, gN3, and gN4), with the gN3 genotype further divided into two subgenotypes (gN3a and gN3b), and the gN4 genotype comprising three subgenotypes (gN4a, gN4b, and gN4c) (9).

CMV is recognized as the most common opportunistic viral pathogen and a major cause of morbidity. Among recipients of hematopoietic stem cell transplantation (HSCT), CMV infection can range from asymptomatic replication to symptomatic end-organ disease (8). Similarly, in kidney transplantation (KT), CMV replication within the graft—whether from host infection or reactivation—remains a leading cause of allograft failure and mortality (11). Approximately 67% of kidney transplant recipients develop CMV disease. In the absence of prophylactic treatment, infection rates can be exceedingly high, reaching up to 100% in the highest-risk groups (12). Although CMV genotyping has been widely investigated globally, particularly in transplant patients, regional data from Jordan remains limited. The genetic distribution of CMV strains in this population is not yet characterized, despite the virus's clinical impact on transplant outcomes. This study addresses a critical gap by investigating CMV prevalence and determining the most common genotypes among HSCT and SOT recipients in Jordan. The findings provide locally relevant insights that may inform regional diagnostic and therapeutic strategies.

MATERIALS AND METHODS

The study design. A retrospective observational study was conducted at the Virology Department of the Jordanian Royal Medical Services (JRMS) Hospital. The study population comprised all patients aged one year or older who had undergone solid organ transplantation (SOT) or hematopoietic stem cell transplantation (HSCT) and were admitted between January and October 2024.

Clinical and demographic data (e.g., age, gender, transplant type) were collected from the medical records. Patients with significant medical conditions at the time of transplantation—such as existing organ failure—that could confound the study outcomes were excluded from the final analysis.

Sample collection. Blood samples were collected in EDTA tubes, plasma was separated at 800-1600×g for 20 minutes, and then stored at -20°C until use.

DNA extraction. CMV DNA was extracted from 200 µL of plasma using the QIAamp Blood Mini Kit (Qiagen, Germany) per the manufacturer's protocol. The purified DNA was stored at -20°C until further use (13).

DNA quantification. DNA concentration and purity were determined using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific). DNA concentration was measured, and purity was assessed based on the A260/A280 ratio. Only samples with an A260/A280 ratio between 1.7 and 2.0 were subjected to PCR (14).

Detection by real-time PCR. CMV DNA was detected by amplifying a specific 105 bp region of the CMV genome using the Artus CMV RG PCR Kit (Qiagen) on a Rotor-Gene Q MDx instrument, according to the manufacturer's instructions. The forward and reverse primer sequences were 5′-GAAGGTGCAGGTGCCCTG-3′ and 5′-GTGTCGACGAACGACGTACG-3′, respectively (15). The PCR was performed in a 50 µL reaction mixture containing DNA template, master mix, and a fluorescent dye. The thermal cycling protocol consisted of an initial denaturation at 94°C for 5 min, followed by 40 cycles of denaturation at 94°C for 15 s, annealing at 65°C for 30 s, and extension at 72°C for 20 s, with a final extension at 72°C for 10 min. Amplification was monitored in real-time, and a sample was considered positive if its cycle threshold (Ct) value was ≤ 30 (commonly 27 ± 3). The quantification standard (QS) range for the assay was 1×10^1 to 1×10^4 copies/µL (13).

Multiplex nested PCR. The gB (UL55) and gN (UL73) genotypes were identified using a multiplex nested PCR technique, as described by Jiang et al. (2017). The first-round PCR was performed in a 20 µL reaction volume using a Quant Gene 9600 thermal cycler (Bioer, China). The reaction mixture for gN genotyping contained the i-StarTaq PCR Master Mix (iNtRON Biotechnology, South Korea), while the i-Taq Maxime PCR PreMix Kit (iNtRON) was used for gB. Each reaction included nuclease-free water, DNA template, and forward and reverse primers (each at 10 µM) (10). The primers for each genotype were selected based on their design in previously published studies (10).

For the second-round PCR, a 20 µL reaction was prepared. For gN genotyping, the i-StarTaq PCR Master Mix was used, while the i-Taq Maxime PCR PreMix Kit was used for gB (both from iNtRON Biotechnology, South Korea). Each reaction contained 0.5 µL of each primer (10 µM), nuclease-free water, and 2 µL of the first-round PCR product as template. A negative control, with nuclease-free water substituted for the template, was included in each run. The cycling conditions for both rounds are summarized in Table 1 (10).

For gB, the second-round amplification was performed over 35 cycles under the following conditions: initial denaturation at 94°C for 2 min; followed by cycles consisting of denaturation at 94°C for 30 s, two-step annealing at 60°C for 1 min (cycles 1-20) and 58°C for 1 min (cycles 21-35), and extension at 72°C for 30 s (cycles 1-20) and 90 s (cycles 21-35); with a final extension at 72°C for 10 min.

For gN, DNA amplification was performed using a two-round nested PCR protocol. The first-round PCR was carried out for 40 cycles under the following conditions: initial denaturation at 94°C for 2 min; followed by cycles of denaturation at 94°C for 30 s, annealing at 57°C for 1 min, and extension at 72°C for 30 s; with a final extension at 72°C for 5 min. The second-round PCR used the first-round product as a template and was performed for 30 cycles using the same cycling profile. The primer sequences and detailed cycling conditions for the multiplex nested PCR are summarized in Tables 2 and 3 (10).

Gel electrophoresis. The PCR products were analyzed by electrophoresis on a 3% agarose gel prepared in 1x TAE buffer and stained with 10 µL of RedSafe nucleic acid stain. Samples were loaded alongside SiZer™-100 and ZR 50 bp DNA ladders as size markers. Electrophoresis was performed at 90 V for approximately 100 minutes, and DNA bands were visualized using a Quantum gel documentation system (10).

Data analysis. Statistical analysis was performed using IBM SPSS Statistics software (Version 25). A p-value of < 0.05 was considered statistically significant. Associations between categorical variables were assessed using the chi-square test.

Ethical considerations. This study was conducted in accordance with the principles of the Declaration of Helsinki. Ethical approval was obtained from the Institutional Review Boards (IRB) of Zarqa University (approval code: IRB/ZU/2024/30) and the Jordanian Royal Medical Services (JRMS) Hospital (approval code: 1544/34/1/11).

Table 1. PCR reaction quantities after standardization for genotyping.

	Reagent	Volume	Conc.
gB	Maxime PCR PreMix Kit (i-Taq)	5 µL	
1st round	Forward primer	1 µL	
	Reverse primer	1 µL	10 µM
	Genomic DNA (sample)	2 µL	10 µM
	nuclease-free water	11 µL	
	The final volume of the PCR tube = 20 µL		
gB	Maxime PCR PreMix Kit (i-Taq)	5 µL	
2nd round	primers	0.5 µL of each primer	10 µM
	Genomic DNA (sample)	1 µL	
	nuclease-free water	11 µL	
	The final volume of the PCR tube = 20 µL		
gN	PCR Master Mix	10 µL	2×
1st round	Forward primer	1 µL	10 µM
	Reverse primer	1 µL	10 µM
	Genomic DNA (sample)	2 µL	
	nuclease-free water	6 µL	
	The final volume of the PCR tube = 20 µL		
gN	PCR Master Mix	10 µL	2×
2nd round	primers	0.5 µL of each primer	10 µM
	Genomic DNA (sample)	1 µL	µL
	nuclease-free water	6 µL	
	The final volume of the PCR tube = 20 µL		

Table 2. gB and gN genotypes and their primer sequences.

Gene	PCR round	Primer	Primer sequence (5′-3′)	Ampilicon size (bp)
gB	1st round	UL55 up	TTTGGAGAAAACGCCGAC	751
		UL55 low	CGCGCGGCAATCGGTTTGTTGTA	
	2nd round	gB1	ATGACCGCCACTTTCTTATC	420
		gB2	TTCCGACTTTGGAAGACCCAACG	613
		gB3	TAGCTCCGGTGTGAACTCC	190
		gB4	ACCATTCGTTCCGAAGCCGAGGAGTCA	465
		gB5	TACCCTATCGCTGGAGAAC	139
		gB low	GTTGATCCACACACCAGGC	
gN	1st round	UL73 up	AGTCGATTCGGTCGGTTAAC	469
		UL73 low	CCACCCTCAATAGCCTTTGGT	
	2nd round	gN1	TTCTGCTAGCGTATCAACTACC	283
		gN2	AGTGCAAAACACTGGTGCT	380
		gN3b	CACAACCACATTAACGAGT	214
		gN4a	CAACAATACGTCGACTGCTAGCACAC	325
		gN4b/c	GACAACTAGTACAACTACGGTGACAA	244
		gN low	GACATTGCTGCTTCCAGAA	

Source: (10).

Table 3. PCR cycling conditions for gB and gN genotyping.

Gene	PCR round	PCR condition	PCR cycle
gB	1st round	ID: 94°C / 2 min	35 cycle
		D: 94°C / 30 sec	
		A: 60°C / 1 min	
		E: 72°C / 30 sec	
		FE: 72°C / 5 min	
	2nd round	ID: 94°C / 2 min	35 cycle
		D: 94°C / 30 sec	
		A: 58°C / 1 min	
		E: 72°C / 90 sec	
		FE: 72°C / 10 min	
gN	1st round	ID: 94°C / 2 min	40 cycle
		D: 94°C / 30 sec	
		A: 57°C / 1 min	
		E: 72°C / 30 sec	
		FE: 72°C / 5 min	
	2nd round	ID: 94°C / 2 min	30 cycle
		D: 94°C / 30 sec	
		A: 57°C / 1 min	
		E: 72°C / 30 sec	
		FE: 72°C / 5 min	

ID: initial denaturation, D: denaturation, A: annealing, E: elongation, FE: final elongation.

RESULTS

Patients' characteristics. Out of all recipients, 80 were identified as CMV-positive. Within the KT group, 15 recipients (18.8%) tested positive for CMV, while a significant majority of the HSCT recipients, 65 (81.2%), were CMV-positive. In the HSCT group, 40 recipients (62%) were male, and 25 (38%) were female. Notably, 54 recipients (83%) were under 18 years of age, while 11 (17%) were above 18. Conversely, in the KT group, there were 9 males (60%) and 6 females (40%), with 3 recipients (20%) under 18 and 12 (80%) above 18. Statistical analysis revealed a significantly higher proportion of females compared to males ($P = 0.044$). Furthermore, recipients under 18 years of age constituted a significantly greater percentage than those above 18 ($p < 0.0001$), as illustrated in Table 4.

Detection of CMV genotypes. Of the 80 CMV-positive samples, 44 had sufficient DNA for genotype analysis; the remaining 36 were excluded due to insufficient quantity/quality. Among the 44 genotyped recipients, 34 (77.2%) were HSCT patients and 10 (22.8%) were KT patients. Within the HSCT group (n=34), 10 (29%) were male and 24 (71%) were female, and 31 (91%) were under 18 years of age. All 10 KT patients were female. Overall, the results indicated a significantly higher prevalence of CMV positivity in individuals under 18 years compared to those over 18, as well as a higher prevalence in females compared to males, as detailed in Table 5.

Detection of CMV gB among HSCT and SOT recipients. Among the 44 CMV-positive samples, 27 were identified as having the gB genotype, with 19 belonging to recipients of hematopoietic stem cell transplantation (HSCT) and 8 to kidney transplant (KT) patients. The identified gB genotypes included gB1, gB2, gB3, and gB4, while gB5 was not detected (Figs. 1A and B).

The gB2 genotype was found in seven samples, representing 25.9% of the CMV DNA-positive cases. The gB4 genotype was compatible with six samples (22.2%), and the gB3 genotype was identified in one sample (3.7%). Additionally, six samples (22.2%) exhibited co-infections with two distinct CMV genotypes, specifically gB1+gB2, gB1+gB4, and gB2+gB3.

Furthermore, seven samples (25.9%) showed a combination of multiple gB genotypes, specifically gB1+gB2+gB4, gB2+gB3+gB4, and gB1+gB2+gB3+gB4. The gB2 genotype was the most prevalent overall, accounting for 31% of cases, and was predominantly observed among HSCT recipients, as presented in Table 6.

Detection of CMV gN among HSCT and SOT Recipients. The gN genotype was identified in 21 out of the 44 CMV-positive recipients. Among these, seven were KT recipients and fourteen were HSCT recipients. The detected gN genotypes included gN1, gN2, gN4a, and gN4b/c, while gN3b was not observed in this group (Figs. 2A and B). Of the 21 gN-positive samples, four (19.05%) contained the gN1 genotype, four (19.05%) the gN2 genotype, three (14.29%) the gN4a genotype, and one (4.76%) the gN4b/c genotype. Additionally, nine samples (42.8%) showed co-infections with two distinct gN genotypes, specifically gN1+gN2, gN4a+gN2, and gN4a+gN4b/c. Among KT recipients, the mixed genotype gN1+gN2 was the most prevalent (42.86%). In contrast, among HSCT recipients, the single genotype gN2 and the

Table 4. Distribution of CMV in HSCT and KT recipients according to age and gender.

Type of transplant	Gender		P value	Age		P value
	Male (N, %)	Female (N, %)		<18 (N, %)	>18 (N, %)	
HSCT	40,62%	25,38%	<0.063	54,83%	11,17%	<0.0001
KT	9,60%	6,40%	<0.439	3,20%	12,80%	<0.02
Total	49,61%	31,39%	<0.044	57,71%	23,29%	<0.0001

HSCT: Hematopoietic stem cell transplant, KT: Kidney transplants, N: number, %: percentage.

Table 5. Distribution of study population according to ages and gender

Transplant type	Gender		P value	Age		P value
	Male (N,%)	Female (N,%)		<18 (N,%)	>18 (N,%)	
KT	0	(10,100%)		0	10	
HSCT	10,29%	24,71%	<0.016	31,91%	3,9%	<0.0001
Total	10	34	<0.0001	31,70%	13,30%	<0.010

HSCT: Hematopoietic stem cell transplant, KT: Kidney transplants, N: number, %: percentage.

mixed genotype gN1+gN2 were the most common, each detected in 21.4% of cases, as shown in Table 7.

Gel electrophoresis results for gB and gN indicated that 11 of the 44 genotyped samples (25%) were negative for both genotypes. This suggests these recipients may harbor other CMV genotypes (e.g., gM, gH, gL, gO). Analysis of the gB and gN genotype distribution revealed that 15 recipients (34%) had mixed infections involving both the UL55 (gB) and UL73 (gN) genes. Of these 15 recipients, 10 (66.7%) were HSCT patients and 5 (33.3%) were KT patients, as presented in Table 8.

DISCUSSION

This is the first study designed to determine the prevalence of CMV infection and its gB and gN genotype among recipients with HSCT and KT in Jordan. CMV is a common opportunistic viral pathogen, frequently causing serious illness and potentially fatal outcomes in HSCT recipients, while in KT recipients, it is linked to a higher risk of transplant rejection (16). CMV genotyping is essential for monitoring treatment outcomes in transplant recipients, as it enables the identification of specific viral strains that may affect antiviral efficacy and disease progression (17).

In our cohort, males represented 60-62% of both KT and HSCT recipients, with the majority of recipients being under 18 years of age. This demographic profile aligns with previous studies reporting a male proportion of 61.1% and a CMV prevalence in children ranging from 24% to 60.6% (10, 16). A key finding of our study was the detection of all four major gB genotypes (gB1–gB4), while the gB5 genotype was absent. Among Jordanian HSCT recipients, the most prevalent genotype was gB2 (31%). This contrasts with reports from other regions. For instance, one study identified gB1 (45%) as the most common genotype in HSCT and KT recipients (18), while another from Serbia found gB4 (38.9%) to be predominant in pediatric HSCT patients (10). This discrepancy in genotype prevalence is likely attributable to variations in the study populations. In our cohort of kidney transplant (KT) recipients, the most common genotypic patterns were mixed infections containing gB2, gB3, or gB4 alongside gB1 or gB2 (25%), and single gB4 infections (25%). This contrasts with previous reports where gB1 was predominant: one study found gB1 in 53.3% of KT recipients (18), and another reported gB1 in 38% of solid organ transplant (SOT) recipients with CMV disease. They found no associations between the various gB genotypes and

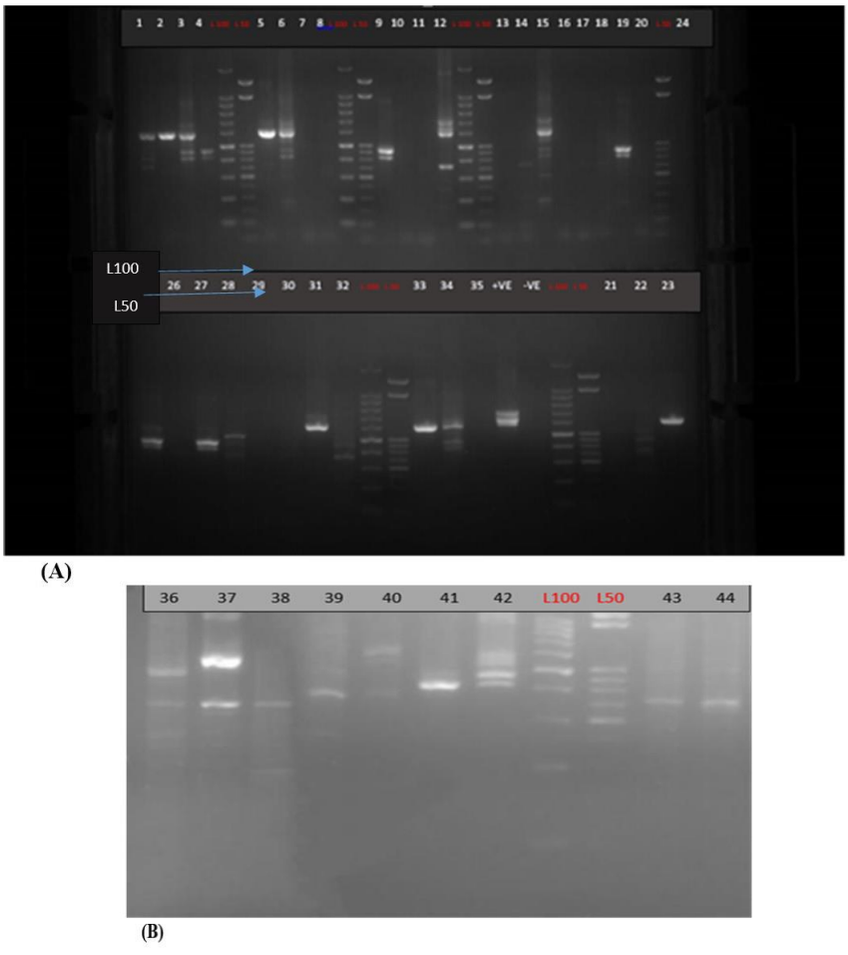

Fig. 1. Agarose gel (3%) electrophoresis of PCR products for gB genotypes 1-5 (lanes 1-44). L100 and L50 represent 100-bp and 50-bp DNA ladders, respectively.

Table 6. Distribution of gB among HSCT and KT recipients

CMV genotype	Total N=27	HSCT N=19	KT N=8
Single genotype	(N,%)	(N,%)	(N,%)
gB2	7 (25.9%)	6 (31%)	1 (12.5%)
gB3	1 (3.7%)	N/A	1 (12.5%)
gB4	6 (22.2%)	4 (21%)	2 (25%)
2 genotypes			
gB1+gB2	1 (3.7%)	1 (5.26%)	N/A
gB1+gB4	4 (14.8%)	4 (21 %)	N/A
gB2+gB3	1 (3.7%)	1 (5.26%)	N/A
3 genotypes			
gB1+gB2+ gB4	4 (14.8%)	2 (10.5%)	2 (25%)
gB2+gB3+ gB4	1 (3.7%)	1 (5.26%)	N/A
All genotypes			
gB1+gB2+ gB3+ gB4	2 (7.4%)	N/A	2 (25%)

HSCT: Hematopoietic stem cell transplant, KT: Kidney transplants, gB: glycoprotein B, N/A- not applicable (no genotypes are detected), N: number, %: percentage.

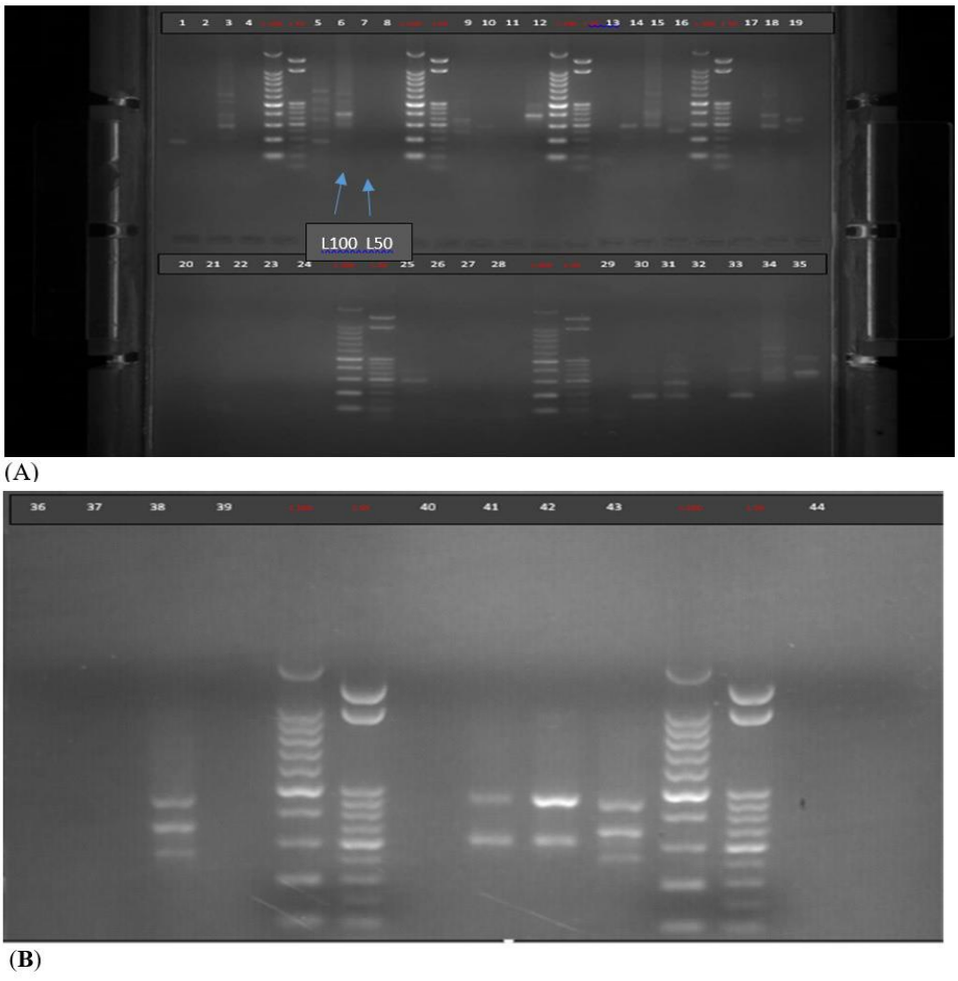

Fig. 2. Agarose gel (3%) electrophoresis of PCR products for gN genotypes 1, 2, 3b, 4a, and 4b/c (lanes 1-44). L100 and L50 represent 100-bp and 50-bp DNA ladders, respectively.

Table 7. Distribution of gN among HSCT and KT recipients.

CMV genotype	Total N=21	HSCT N= 14	KT N= 7
Single genotype	(N,%)	(N,%)	(N,%)
gN1	4 (19.05%)	2 (14.29%)	2 (28.57%)
gN2	4 (19.05%)	3 (21.4%)	1 (14.29%)
gN4a	3 (14.29%)	3 (21.4%)	N/A
gN4b/c	1 (4.76%)	1 (7.14%)	N/A
2 genotypes			
gN1+gN2	6 (28.57%)	3 (21.4%)	3 (42.86%)
gN4a+gN2	1 (4.76%)	N/A	1 (14.29%)
gN4a+gN4b/c	2 (9.52%)	2 (14.29%)	N/A

HSCT: Hematopoietic stem cell transplant, KT: Kidney transplants, gN: glycoprotein N, N: number, %: percentage.

acute graft rejection in kidney or liver transplant recipients, nor did they find any differences between genotypes in terms of the development of clinical disease or CMV load (19, 20).

Furthermore, regarding the gN genotype, we observed the four major genotypes (gN1, gN2, gN4a, and gN4b/c), with the notable absence of gN3b. This finding contrasts with previous studies, which reported the absence of different genotypes, such as gN2, while detecting others (21). In our study, the distribution of gN genotypes among HSCT recipients showed that gN2, gN4a, and the mixed genotype gN1+gN2 were equally prevalent (21.4% each). In contrast, among KT recipients, the mixed genotype gN1+gN2 was the most prevalent (42.8%). This differs from other studies on pediatric HSCT, which reported the detection of five gN genotypes, with gN1

Table 8. Distribution of mix genotype among HSCT and KT recipients.

Mix genotypes					
Number	**KT**		**Number**	**HSCT**	
	5 (33.4%)			**10 (66.6%)**	
1	gB1,gB2,gB4	gN1,gN2	1	gB2	gN4a
2	gB1,gB2,gB4,gB3	gN2	2	gB1,gB4	gN4b/c,gN4a
3	gB1,gB2,gB4,gB3	gN1	3	gB3,gB2	gN2
4	gB1,gB4,gB2	gN4a, gN2	4	gB3,gB2,gB4	gN4b/c,gN4a
5	gB3	gN1,gN2	5	gB1,gB4,gB2	gN1
			6	gB4	gN1,gN2
			7	gB2	gN1,gN2
			8	gB2	gN2
			9	gB1,gB4	gN4a
			10	gB1,gB4	gN4a

HSCT: Hematopoietic stem cell transplant, KT: Kidney transplants, gN: glycoprotein N, gB: glycoprotein B

being the most common (34.6%) (10). The gN1 genotype has been reported as the most prevalent in solid organ transplant (SOT) recipients and in patients with acquired immunodeficiency syndrome (AIDS) (10), while another study found gN4a (49.2%) to be most prevalent among AIDS patients (8). The variance in reported genotypes is likely due to the limited research on gN in transplantation and differences in study populations.

Notably, 15 recipients (34%) presented with mixed genotypes at both the UL55 (gB) and UL73 (gN) loci. Of these, 10 (66.7%) were HSCT recipients and 5 (33.3%) were KT recipients. This successful dual-locus genotyping contrasts with some previous studies where one or both genotypes could not be determined in all cases (10). Eleven of the 44 genotyped samples (25%) were negative for both gB and gN. This suggests these recipients may harbor other CMV glycoprotein genotypes, such as gM, gH, gL, or gO. Further studies are needed to confirm this hypothesis and determine its generalizability.

Our main limitations are typically related to those of retrospective observational studies, in which routinely collected electronic health record data are used, with some missing data, residual confounders, and potential biases. This study was conducted at a single center (JRMS Hospital); therefore, its findings may not fully reflect the prevalence of CMV among HSCT and SOT recipients across Jordan. Furthermore, our analysis was limited to the detection of gB and gN genotypes using specific primers. Other clinically relevant genotypes, such as gM, gH, gL, and gO, were not identified in this study. However, we recommend further studies with a larger sample size and a broader range of data to enable more robust correlations, as well as the use of whole-genome sequencing to better characterize the diversity of CMV strains in HSCT and SOT patients.

CONCLUSION

CMV is a common opportunistic pathogen that poses serious health risks to transplant recipients. In HSCT patients, CMV infection can be life-threatening, while in KT recipients, it significantly increases the risk of graft rejection. Based on our findings, the most prevalent genotypes were gB2 among HSCT recipients and gN1+gN2 among KT recipients. These genotype-specific patterns could influence disease severity and therapeutic responsiveness. Therefore, large-scale studies are needed to map genotype distribution and evaluate its utility in guiding CMV prophylaxis and personalizing treatment for transplant recipients.

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