QUESTION 66
The investigation most useful in the management of cytomegalovirus (CMV) disease in the immunocompromised host is:
A. CMV blood culture.
B. direct CMV antigen detection in peripheral blood mononuclear cells (PBMCs).
C. CMV immunoglobulin M (IgM).
D. plasma CMV DNA concentration (viral load).
E. early antigen detection in supernatant after culture of PBMCs for 48 hours.


Panel 1. Diagnosis of cytomegalovirus infection and disease
History/examination
Presence of cytomegalovirus in tissue might simply be a bystander effect and does not necessarily imply causality. Cytomegalovirus-associated disease should be diagnosed on clinical grounds in combination with detection of virus.

Viral cultures
Requires at least 21 days to be reported negative. Not routinely done, but can be useful in resistance testing.

Detection of early antigen fluorescent foci
24h turn-around. Infected fibroblasts stained with fluorescent antibody specific for the antigen MIE p72. Insufficiently sensitive to be routinely recommended after allogeneic stem-cell transplantation.

Antigenaemia assay
Quantitates leucocytes positive for pp65. Since uninfected cells can harbour cytomegalovirus protein, provides an indirect assessment of infection. Reliable, rapid, and sensitive. In common use.

Qualitative PCR
Can be used on whole blood, leucocytes, and plasma. Rapid and sensitive, easily automated. Threshold needs careful calibrating to over detection of cytomegalovirus infection.

Quantitative PCR
Allows response to treatment to be monitored. Useful as a surrogate marker of clinical or viral resistance. Correlation between high degrees of viral DNA and presence of clinical symptoms in transplant recipients and patients with AIDS and the congenital infection of pregnancy has been shown.

Hybrid capture assay
RNA probes used to detect viral DNA in an ELISA-type format. Can be used on whole blood stored for up to 48 h. Limited experience but results to date promising.

Nucleic assay sequence-based amplification
Allows the specific nucleic assay sequence-based amplification of unspliced viral mRNAs in a background of DNA. Experience in transplant and AIDS patients encouraging.

Panel 2. Monitoring of cytomegalovirus-specific immunity
Serology
Highly specific and sensitive in immunocompetent individuals. IgM titres rise 2–6 weeks after infection and can persist for 2 years and be detectable during episodes of reactivation. IgG seropositivity is usually lifelong although antibody concentrations can decline with age. Unreliable and not used in monitoring of cytomegalovirus reactivation in immunocompromised patients. IgG antibody avidity assays (low avidity suggests infection within 3 months) can be useful in identifying pregnant women at risk of transmitting intrauterine infection.

Tetramer assays
Remains a research method to detect peptide-specific CD8+ T-cells. Data in allogeneic stem-cell transplant setting suggest a protective threshold below which patients at high risk of reactivation can be identified.\textsuperscript{72} and \textsuperscript{73}

**Cytokine assays**

Research method to enable high throughput detection of virus-specific T-cells. Being assessed in transplant patients. As with tetramer assays, when combined with sensitive, rapid viral detection techniques, may be of use in monitoring post-transplant patients and to assist decision-making in pre-emptive or prophylaxis strategies.

**Answer:** D. plasma CMV DNA concentration (viral load).