Sudan University of Science and Technology
College of Graduate Studies

Seroprevalence Of Cytomegalovirus Infection among Kidney Transplant Recipients at East Nile Model Hospital Khartoum State

الانتشار المصلي للإصابة بفيروس مضخم الخلايا بين متلقي الكلي بمستشفى شرق النيل النموذجي في ولاية الخرطوم

A thesis submitted to the College of Graduate Studies in Partial fulfillment for the requirement of MSc. Degree in Medical Laboratory Science (Microbiology)

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Principle of Human IgG CMV ELISA

The HUMAN CMV IgG ELISA is based on the classical ELISA technique. The microtiter strip wells as solid phase are coated with cell culture derived CMV antigens (CMV Ag). In the first incubation step corresponding specific antibodies (CMV-IgG-Ab) present in patient specimens or controls bind to the antigens at the solid phase. At the end of the incubation unbound components are washed out. For the second incubation step anti- IgG conjugate (anti- human IgGb antibodies, peroxidase conjugated) is added which binds specifically to IgH antibodies resulting in the formation of typical immunocomplexes. After a second washing step to remove unbound conjugate, TMB/Substrate is added (step 3). A blue color is directly proportional to the CMV-IgG Ab concentration in the specimen. The absorbance of controls and specimens is determined by using ELISA microplate reader (human Reader). Results for patient samples are obtained by comparison with cut-off value.

Principle of Human IgM CMV ELISA

The HUMAN CMV IgM ELISA is based on the classical ELISA technique. The microtiter strip wells as solid phase were coated with cell culture derived CMV antigens (CMV Ag). In the first incubation step corresponding specific antibodies
(CMV-IgM-Ab) present in patient specimens or controls bind to the antigens at the solid phase. The sample dilution buffer contains anti-human IgM to prevent rheumatoid factor (RF) interference and competition for specific IgM present in the specimen.

At the end of the incubation, unbound components are washed out. For the second incubation step anti-IgM conjugate (anti-human IgM antibodies, peroxidase conjugated) is added which bind specifically to IgG antibodies resulting in the formation of typical immunocomplexes. After a second washing step to remove excess conjugate, TMB/Substrate is added (step 3). A blue color develops changing to yellow after stopping the reaction. The intensity of the color is directly proportional to the CMV-IgM-Ab concentration in the specimen.

The absorbance of controls and specimens is determined by using ELISA micro plate reader (human Reader). Results for patient samples are obtained by comparison with cut-off value.
REFERENCES