Detection of Cytomegalovirus Antibodies by two Serological Immunoassay techniques

Thesis

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by

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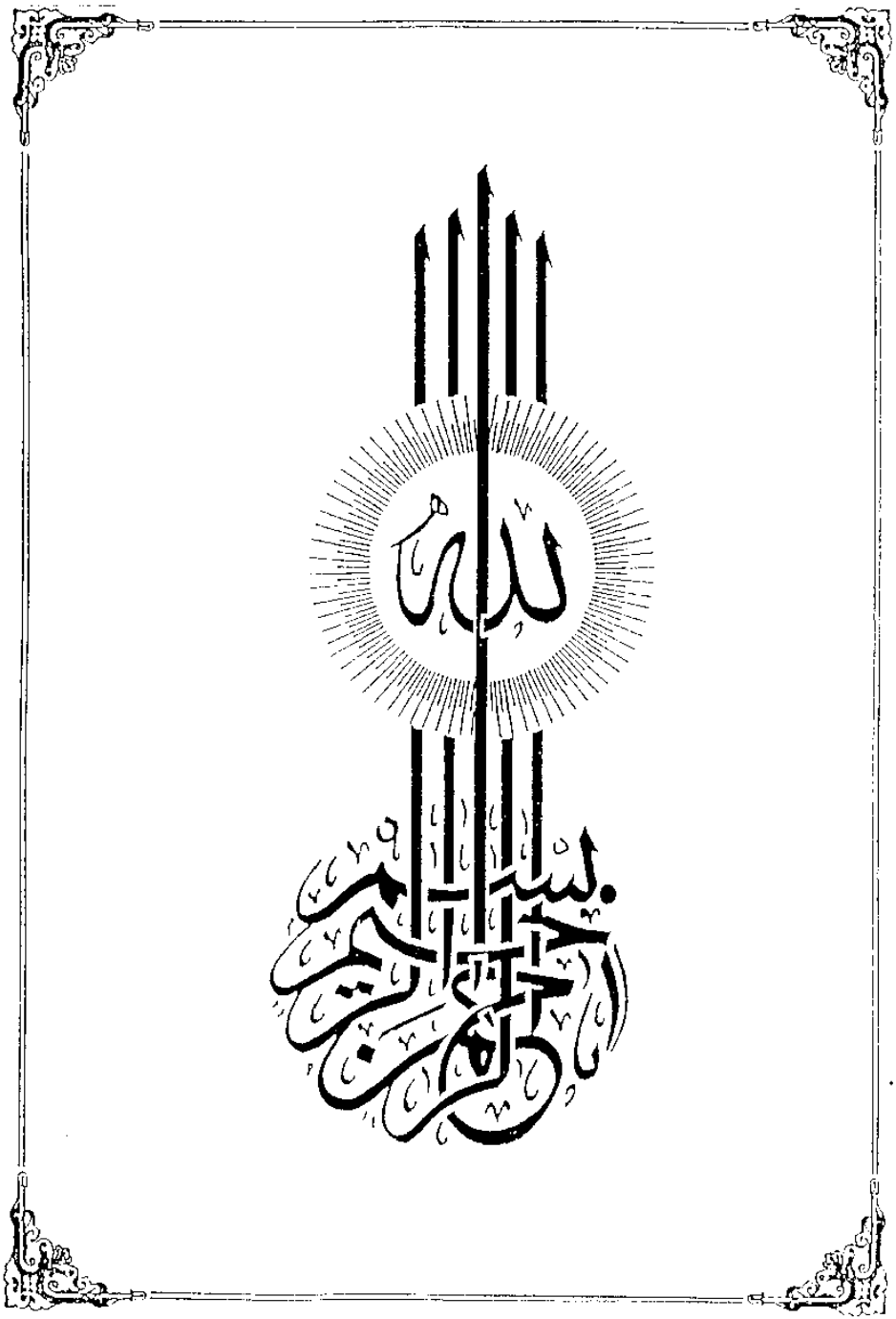
وقل اعملوا فسيرى الله عملكم ورسوله والمؤمنون

صدق الله العظيم



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INTRODUCTION

Cytomegalo virus (CMV) is a member of the Herpes viridae. As with other herpes viruses, Primary CMV infection is often followed by persistent and/or recurrent infection. The latter are most often caused by reactivation of latent virus but reinfection also occurs possibly because of antigenic diversity of CMV *(Alford & Britt 1990)*.

CMV infections are rarely symptomatic in immunological competent individuals *(Bayer et al., 1984)*, but there is a risk of serious complications when CMV infections occur congenitally or in immunocompromised individuals *(Leland et al., 1989)*.

CMV seropositive donors of blood and transplanted organs are an important source of CMV infection in their recipients *(Ho, 1982)*.

Detection of serum antibody can help in identifying those individuals with prior CMV exposure and may aid in confirming acute CMV infection. More over absence of CMV antibody by serological testing can identify those individuals at risk for CMV infection. This extremely important in immunocompromised patients who may receive blood products and based on CMV seronegativity, should be given seronegative CMV blood products *(Bowden et al., 1987)*.

CMV antibodies can be measured by a variety of serological immunoassays as complement fixation, solid phase radioimmunoassay, indirect hemagglutination assay and enzyme-linked immunosorbent assays (ELISA) *(Bowden et al., 1986)*.

Aim of Work

The aim of this study is to compare (ELISA and CF techniques) to find out which is more sensitive as regard the detection of CMV antibody. This will help for selection of this sensitive technique for detection of CMV infection in (e.g) donors and recipients of organ transplantation.

REVIEW of LITERATURE

A- Herpes Viridae

Herpes viruses of human include herpes simplexvirus type 1 and 2, varicella zoster virus, cytomegalovirus, and Epstein-Barr (EB) virus. Anew herpes virus, human B-lymphotropic virus (HBLV), has been recovered from patients with lymphoproliferative disorders. It has been designated human herpes virus 6. *(Jawetz et al., 1989)*

1. Characteristics of Herpes Viruses

Herpes virus virions consist of :

a) An envelope with surface projections.
b) A tegument consisting of a morphous material.
c) An icosahedral nucleocapsid, 100nm in diameter with (162) prismatic capsomeres.
d) A core consisting of a fibrillar spool on which the DNA is wrapped.Virus particles have an overall diameter of (150 to 220) nm. The genome consists of one molecule of ds DNA, size (120 kbp to 220 kbp) . Genomic DNA contains terminal and internal reiterated sequences, usually forming two covalently linked components (L&S), arranged in five different patterns. Virions have more than (30) structural proteins including in some cases an FC receptor in their envelope *(Murphy. 1988)*

2. Classification

Classification of the numerous members of herpes virus family is complicated. A useful division into sub-families is based on biologic properties of the agents.

a) Alpha herpes viruses are fast growing, cytolytic viruses that tend to establish latent infections in neurons.
b) Beta herpes viruses are slow growing and cytomegalic (massive enlargement of infected cells) and become latent in secretory glands and kidneys.
c) Gama herpes infect lymphoid cells *(Jawetz et al., 1989)*.
d) A new herpes virus, human B-lymphotropic (HPLV), has been recovered from patients with lymphoproliferative disorders. It has been designated human herpes virus 6 *(Jawetz et al., 1989)*. Classification of human herpes viruses is shown in (Table 1).

There is a little antigenic relatedness among members of herpes virus groups. Only herpes simplex type (1&2) share a significant number of common antigen. This is not surprising, since there is approximately (50%) homology between those two viral genomes *(Jawetz et al., 1989)*.

3) Human Pathogens

a) Simplex Virus : herpes simplex virus (1&2), cercopithecine herpes virus 1 (B-Virus).
b) Varicellovirus : varicella-zoster virus.
c) Cytomegalovirus : human cytomegalo virus.
d) Lymphocrypto Virus : E B virus *(Murphy, 1989)*

Classification of human herpes viruses (*Jawetz et al., 1989*).

Subfamily	Biologic properties		Examples		
	Growth cycle	Cytopathology	Latent infection	Official name	Common name
Alpha herpes virus	Short	Cytolytic	Neurons	Human herpes virus 1.	Herpes simplex virus type-1.
				Human herpes virus 2.	Herpes simplex virus type-2.
				Human herpes virus 3.	Varicella zoster virus
Beta herpes virus	Long	Cytomegalic	Glands kidneys	Human herpes virus 5.	Cytomegalovirus
Gamma herpes virus	Variable	Lymphoproliferative	Lymphoid tissue	Human herpes virus 4.	Epstein Barr (EB) virus

Table 1.

4) Animal Pathogens

a) Simplex Virus : infectious bovine rhinotracheitis virus, bovine mammillitis virus, cercopithecine herpes virus 1 (B-virus) & 2.

b) Varicellao virus : Pseudo rabies virus (of swine), equine rhinopneumonitis and coital exanthema viruses.

c) Muromegalo virus : murine cytomegalo virus.

d) Lymphocrypto virus : baboon herpes virus, pongine(chimpanzee) herpes virus.

e) Thetalymphocrypto virus : Marek's disease herpes virus(of fowl) turkey herpes virus.

f) Rhadino Virus : herpes virus ateles, herpes virus saimiri *(Murphy, 1989).*

B- Cytomegalovirus

1. History

The history of cytomegalovirus began with the discovery of its anatomic pathogenic effect. This preceded by more than (70 years) the modern era initiated by the isolation of the virus. In 1881 Ribbert *(Ribbert, 1904)* first noted large "Protozoan-like" cells in the kidney of a stillborn. By (1932), 25 cases of cytomegalic inclusion disease were described. *(Farber & Wolbach, 1932)* found by postmortem examination that (12%) of the submaxillary glands from (183) infants who died of various causes had intranuclear and cytoplasmic inclusions, suggesting that the infection was not a rare event.

The modern virologic era began with the isolation of murine cytomegalovirus in mouse cell culture by *(Smith,1954)*. Later the isolation of human cytomegalo virus was accomplished independently by *(Smith 1956, &Weller et al., 1957)*.

The term "cytomegaloviruses" was coined by *(Weller et al., 1960)* to replace"Salivary gland virus" or "Cytomegalic inclusion disease virus" *(Klemola and kaarianen 1965)* first described recognizable CMV infection and disease in a normal, healthy adult.

2. Morphology

Cytomegalo virus is the largest member of the human herpes virus family. Physically CMV is approximately (200 nm) in diameter, making it one of the largest animal viruses. The virus consists of 64