## DETECTION OF REACTIVATION OF CYTOMEGALOVIRUS IN RENAL TRANSPLANT RECIPIENTS

### **Thesis**

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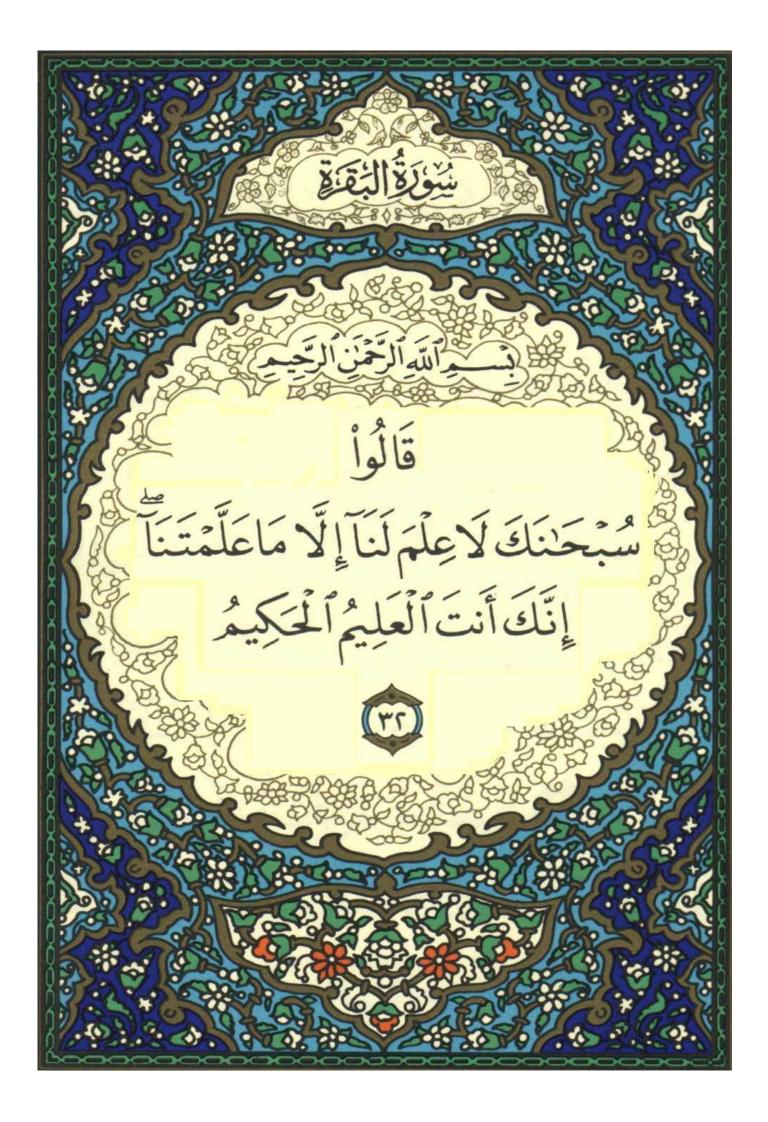
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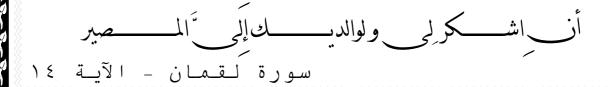
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### Abstract

**Background:** Cytomegalovirus infection in renal transplant recipients is a major clinical problem that may cause significant morbidity and mortality. Infection can occur as a result of reactivation of latent virus or new infection from donor tissues.

*Objectives:* To assess the incidence of cytomegalovirus (CMV) reactivation, and to determine the predictive factors for CMV reactivation in renal-transplant patients also to compare CMV-DNA amplification using qRT-PCR with serologic assays of CMV-IgM antibodies to detect CMV reactivation.

Study design: Sixty patients were included in this study. They were classified into 3 groups based on the post transplantation period during which the study was performed. ELISA was used to detect the pre-transplantation CMV serostatus for the donor and the recipient as well as the recipient post transplantation CMV serology. CMV DNAemia was assessed by qRT-PCR first on whole blood (WB). Whenever a positive result was obtained; the assay was then performed on plasma to detect the difference between them.

**Results:** CMV reactivation occurred in two patients following the treatment of their rejection episode and was detected by qRT-PCR using whole blood and not in plasma.

Conclusion: Cytomegalovirus reactivation was not high in the studied patients, which may be due to the presence of pre -existing immunity in the form of neutralizing antibody, and that treatment of an episode of acute allograft rejection was the most important risk for CMV reactivation within the first year post-transplantation. qRT-PCR is an important tool in predicting subsequent or ongoing disease, while detection of anti CMV-IgM antibodies is not sensitive enough for diagnosis

*Keywords: Cytomegalovirus*, Kidney Transplantation, Reactivation, Acute rejection, Immunosuppressive therapy.

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# List Of Abbreviations

Ab Antibody

ADCC Antibody-dependent cell-mediated cytotoxicity

Ag Antigen

AIDS Acquired immune deficiency syndrome

ALG Antilymphocytic globulin

ALL Acute lymphoblastic leukemia

AP Assembly protein

BKV BK virus

BMT Bone marrow transplantation

CARI Caring for Australians with Renal Impairment

CD Clusters of differentiation

CDV Cidofovir

CMV Cytomegalovirus

CNS Central nervous system

CP crossing point

CPE Cytopathic effect

CsA Cyclosporin A

C<sub>T</sub> Threshold cycle

CTL Cytotoxic T-cell

CVS Cardiovascular system

D/R Donors/recipients

DB Dense bodies

DM Diabetes Mellitus

DMSO Dimethyle sulfoxide

DNA Deoxyribonucleic Acid

dNTP desoxynucleoside triphosphate

dTTP desoxythimidine triphosphate

dUTP desoxyuridine triphosphate

EBV Epstein-Barr virus

EDTA Ethylene-diamine-tetraacetic acid

EHV1 Equine Herpesvirus

ELISA Enzyme-linked immunosorbent assay

ER Endoplasmic reticulum

ESRD End stage renal disease

FRET Fluorescence resonance energy transfer

gB gene Glycoprotein B gene

GC Guanine & Cytosine

GCV Gancyclovir

H&E Hematoxylin and eosin, a standard tissue stain

H<sub>2</sub>O Water

H<sub>2</sub>O<sub>2</sub> Hydrogen peroxide

H<sub>2</sub>SO<sub>4</sub> Sulpheric acid

HAART Highly active anti-retroviral therapy

HBV Hepatitis B Virus

HCMV Human cytomegalovirus

HCV Hepatitis C Virus

HD Hemodialysis

HHV Human herpesvirus

HHV-8 Human herpesvirus 8

HIV Human Immunodeficiency Virus

HLA Human leukocyte antigen

Hr Hour

HRPO Horse radish peroxidase

HSCT Haematopoietic stem cell transplant

HSV Herpes Simplex Virus

HVS Herpesvirus Saimiri

IE genes Immediate early genes

IF Immunofluorescence

IgA Immunoglobulin A

IgG Immunoglobulin G

IgM Immunoglobulin M

IHMF The International Herpes Management Forum.

INF( $-\alpha$ ) Interferons  $-\alpha$ 

IU International unit

Kbp. Kilo base pair

KS Kaposi's sarcoma

L genes Late genes

Lab Laboratory

LED Light emitting diode

MGPs Magnetic glass particles

MHC Major histocompatibility complex

MIE gene Major immediate-early gene

ml Milliliter

MMF Mycophenolate mofetil

mmol/L Millimols per litre

NK cells Natural killer cells

 $N^{\underline{o}}$  Number

°c Degree Celsius

OKT3 Orthoclone

PBLs Peripheral blood leukocytes

PBMCs Peripheral blood mononuclear cells

PCR Polymerase chain reaction

PD Peritoneal dialysis

PFA Pyrophosphate analogue

pp Phosphoprotein

PRV Pseudorabies virus

PVAN Polyomavirus-associated nephropathy

Qn PCR quantitative PCR

RBC Red blood cells

RNA Ribonucleic acid

RPR Rapid plasma regain

RT- PCR Real-time polymerase chain reaction

RTR Renal transplant recipients

SCT Stem cell transplant

SE Standard error of the mean

SOT Solid organ transplant

SPSS Statistical Product and Service Solutions (formerly Statistical

Package for Social Sciences).

TMB Tetramethyl benzidine

TST Tuberculin skin test

UL Unique long

US Unique short

UTI Urinary tract infection

vICA Viral inhibitor of caspase-8 induced apoptosis

vMIA Viral mitochondrial inhibitor of apoptosis

WB Whole blood

WBC White Blood Cell, White Blood Cell Count

WHO World Health Organisation

μg Microgram

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# INTRODUCTION INTRODUCTION AND SOFT WORK AND