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Molecular Detection of Human Herpes virus type 7 (HHV-7) among Renal Transplant Patients in Khartoum State, Sudan

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ABSTRACT

Background:

Human herpesvirus 7 (HHV-7) is one of eight known members of the Herpesviridae family that infects humans. HHV-7is a member of Betaherpesviridae, a subfamily of the Herpesviridae that also includes HHV-6 and Cytomegalovirus (HHV-5 or HCMV).[1] HHV-7 often acts together with HHV-6, and the viruses together are sometimes referred to by their genus, Roseolovirus. HHV-7 was first isolated in 1990 from CD4+ T cells taken from peripheral blood lymphocytes. This study was carried out to detect the HHV-7 and virus nuclic acid among renal transplant patients in Khartoum state.

Method:

This was descriptive study in which serum specimens were collected from 95patient(72male,27 female)- and investigated for HHV-7 DNA using Nested PCR The study group age ranged from 20 to 80years old.

Results: All the samples were negative for HHV-7.

Conclusion:

HHV-7 was not documented in renal transplant patients. Further study should be done.

Keywords: Human herpes Viruses type 7(HHV-7).

I. INTRODUCTION

Human herpes virus type 7 (HHV-7) is one of eight known members of the Herpesviridae family that infects humans. HHv- 7 is a member of Betaherpesviridae, a subfamily of the Herpesviridae.

HHV-7 is very similar to that of HHV-6, although it is about 10% smaller,[4] with a DNA genome of about 145,000 base pairs.[5] There are a number of key differences between the genome of HHV-7 and that of HHV-6, but the importance of them for viral DNA replication is not yet known[5]

Both HHV-6B and HHV-7, as well as other viruses, can cause a skin condition in infants known as exanthema subitum, although HHV-7 causes the disease less frequently than HHV-6B.[6] HHV-7 infection also leads to or is associated with a number of other symptoms, including acute febrile respiratory disease, fever, rash, vomiting, diarrhea, low lymphocyte counts,[7] and febrile seizures,[8] though most often no symptoms present at all. ease. Multivisceral involvement with HSV infection is often fatal.

Over 95% of adults have been infected and are immune to HHV-7 and over three quarters of those were infected before the age of six. Primary infection of HHV-7 among children generally occurs between the ages of 2 and 5, which means it occurs after primary infection of HHV-6.[6]

II. MATERIAL AND METHODOLOGY

Study area:

The study were conducted in Khartoum State during the period November 2015 to June 2016.

Inclusion Criteria Patients and Sample:

Patient who were suspected (renal transplanted patient) of acquiring HHV-7infection after kidney transplantation were included in this study.

Blood Sample Collection:

Blood samples were collected from 95 renal receipient paitents (with age ranging from 20 to80years old)-. Blood (2-5 ml) was collected in plain containers and transported on wet ice to the laboratory for immediate processing. Sera were separated from blood samples by- centrifugation for 5 mints at 3000 RPM. Obtained sera were used DNA extraction for Nested PCR.

DNA EXTRACTION:

(QLAamp DNA, Qiagen ,Germany)according to the manufacturers instruction. was extracte from virus samples using Vivantis GF-1 Nuclic acid extraction kits,

Nested PCR:

Nested PCR involve the use of twe primer sets and two successive PCR reaction . the first set of primers and designed to anneal to sequences upstream from the second set of primers and are used in an initial PCR reaction. Amplicons resulting from the first PCR reaction are used as template for second set of primers and second amplification step.

Standard and nested PCRmix contained 1x Taq Polymerase buffer, 1.5 to 2.5mM MgCl2 (MgCl2 concentration was optimized for every PCR)200 μM deoxynucleotide triphosphate (dNTP), 2.5U of TaqPolymerase (all from Thermo Fisher Scientific, Waltham, MA), and 200nM of each primer (IDT Technologies). Forstandard PCR, 100 ng of DNA from samples or (equivalent to thenumber of moles contained in 100 ng of genomic DNA) were used.

In order to run the amplification reactionunder identical mass/volume DNA concentration for samples. For the nested PCR, 0.5 or 0.05μL(1 : 100 or 1 : 1000 dilution resp.) of product of first roundPCR was used as template. All PCR reactions were carriedout in a final volume of 50 μL. All primer sequences andcycling conditions used are detailed in Table1. PCRproducts were analyzed by electrophoresis in 1.8% agarose gels stained with ethidium bromide and photographed under ultravioletlight using the Quantum ST4 System (VilberLourmat, Torcy,Marne-la-Vall´ee, FR). The viral identity of PCR amplifiedfragments was confirmed by sequencing.

Table1. PCR cycling conditions and primers sequences.

virus	Type of PCR	Cycling condition	Primers sequences(5-3)
HHV-7	Standard	95C/45sec,53C/45sec,72C/45sec(30x)	**F**TTTTTACATTTGGCTTGCTTTTTG **R**ATATTTCTGTACCTATCTTCCCAA {56}(c)
HHV-7	Nested	95C/30sec,55C\30sec,72C/30sec(15x)	**F**GAACGGTTTGCTTAGATTGC **R**GCAGACCAAACTCCACAAATTC(b)

a-All amplification runs included an initial denaturation step at 95◦C for 5 minutes and a final extension step at 72◦C for 10 minutes. Annealing temperatures

were optimized for every reaction.

(b)These primers were designed in our laboratory using the Primer-BLAST program [25].

(c)The specificity of the previously reported primers was corroborated using Primer-BLAST program [25].

III. RESULT

A total of 95 renal transplant paitents were enrolled in this study . Their ages ranged from 20 to 80 years, The results were Negative for HHV-7 by Nested PCR.

IV. DISCUSSION

The aim of this study to detection HHV-7 among renal transplant recipients in Khartoum State using Nested PCR .

The result showed that HHV7 were negative 0(0%) in the tested sample.

adult renal transplant recipients at Washington University/Barnes Hospital, The purpose of this study was to compare the prevalence of human herpesvirus (HHV)–7and viremia and the effects of oral and intravenous (iv) ganciclovirin renal transplant recipients . Stored lysates from peripheral blood leukocytesfrom 92 patients, who had been previously analyzed for by polymerase chainreaction (PCR) for 12 weeks after transplantation, for HHV-7 viremia. Baselineand peak prevalences of HHV-7 viremia and 54%,(P ! .0001). (89%) of 92 patients had at least 1 positive PCR forHHV-7. Oral ganciclovir and treatment with iv ganciclovir had no effect on the prevalence of HHV-7 viremia. In contrast These results indicate that HHV-7 .

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