HSV 2 IgG, IgM, ANTIBODY MARKERS IN HIV / AIDS PATIENTS

Jeyakumar Williams*, Prasanna G**, Thirunavukkarasu D***

SUMMARY

Thirty – three males, 47 females were the study materials for our study. Type specific ELISA test for HSV2 IgG and HSV2 IgM were performed in These 80 patients. Sixteen patients shwoed HSV2 IgG, HSV2 IgM markers. Seventy-five per cent HSV2 antibody positives were asymptomatic 12.5 per cent symptomatics were HSV2 IgG positives 12.5 per cent symptomatics were HSV2 IgM positives. One female adult AIDS patient had shown herpetic ulcers in the antrum of the stomach by endoscopy.

Like HSV 1/HIV hybrids, there is a possiblity of HSV2 / HIV hybrids. HIV/HSV2 hybrids could not be made out in this short study. For want of facilities, culture PCR, antigen, capture assays could not be done. Large - scale of study should be done to bring out the clinical variations and therapeutic efficacy in this co-infection in future.

KEY WORDS

HSV2 IgG, IgM, HIV / AIDS, GMKMCH, SALEM

INTRODUCTION

Herpes simplex virus and resulting infections were first identified and demonstrated in early 1900\textsuperscript{1}. It is one of the major opportunistic infections associated with AIDS\textsuperscript{2}. In India also, genital herpes is a major factor in the current HIV 1 epidemic\textsuperscript{3}. HSV 2 and its potential interaction with HIV have emerged as a major public health problem for countries facing the global HIV 1 pandemic\textsuperscript{4}. The outstanding property of herpes virus is their ability to establish life long persistence in their host and to undergo periodic reactivation. The degree of immunosuppression appears to influnce reactivation and severity of disease\textsuperscript{5}.The prevalence of HSV2 shedding was nearly four times greater in HIV seropositives with increased risk of transmission of HSV in the presence of genital ulceration\textsuperscript{6}. Most patients with genital herpes are unaware of their infection status, partly because of the disease can be mild or mistaken, for other syndromes by clinicians and patients. The study of HSV2 and HIV 1 infections in India add important component to find out the interaction between these two viral infections. Type specific serological tests are useful in confirming the diagnosis of the genital herpes and to diagnose persons with unrecognized (non primary and latent) infections\textsuperscript{7}. Oral Acyclovir promptly reduces the frecuency of reactivations of HSV. Primary prevention of HSV in young attendees of STD Clinics could have a major role in the prevention of HIV acquisition.

MATERIALS & METHODOLOGY

Thirty—Two HIV seropositive adult males, one AIDS adult male, 45 HIV seropositive adult females, one AIDS adult female, and one HIV seropositive female child were selected for our study. They were all attendees of Out Patients, Department of STD, Government Mohan Kumar Mangalam Medical College Hospital, Salem.

All the patients except the child were counseled about this study in a separate room confidentially. We have got the consent from the Ethics Committee for conducting this study. We have also obtained the informed consent from the patients.

Period of Study : July 6, 2004 to July 6, 2005.

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TESTIGATIONS
URINE: Albumin, Sugar, Deposits.
Only in 7 patients, we have ENDOSCOPY who showed symptoms of dysphagia and odynophagia.
"Il the details were collected and analyzed.

METHODOLOGY
Screening of HSV2 IgG & IgM antibodies were performed by ELISA technique using "GLOBE DIAGNOSTIC" S.Ol.20096. Seggiono di Piocella (Adiram) - ITALY.

PRINCIPLE OF ASSAY
Microtitre plates were coated with HSV II antigen (Commercially purchased). The solid phase is first treated with diluted sample of the patients. After the washing step, the antibodies specifically bound to antigen are detected with Goat anti-human antibodies conjugated with Horse Radish Peroxidase (HRP).
The substrate / chromogen TMB (Tetra methyl benzidine) solutions is added and the intensity of the generated colour is proportional to the amount of the HSV antibodies in the samples.

ASSAY PROCEDURE FOR HSV 2
All the sera were stored at -20°C. One hour before testing, all the patients sera, all reagents and controls were brought to room temperature and are mixed to vortex mixture. The samples were pre-diluted with specific diluent (sample diluent) by mixing 10u1 with 500u1 diluent in a marked dilution tube.

ELISA PROCEDURE
After dilution of the patients samples, the tests were carried out as per manufacturer’s instructions.

Immediately the absorbance of each well at 450 nm and 620-630 nm was taken by ELISA reader (BIO-RAD ELISA READER).

ADVANTAGES
1. Serodiagnosis is often more rapid than virus isolation.
2. Least expensive way to establish a laboratory diagnosis.
3. Very Useful in ruling out a particular viral diagnosis.

EPIDEMIOLOGICAL PROFILE OF 80 HIV / AIDS PATIENTS

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Subject</th>
<th>Males</th>
<th>Females</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Sex</td>
<td>33</td>
<td>47</td>
</tr>
<tr>
<td>2.</td>
<td>Age</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0-10 years</td>
<td>0</td>
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</tr>
<tr>
<td></td>
<td>11-20 years</td>
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<td>2</td>
</tr>
<tr>
<td></td>
<td>21-30 years</td>
<td>8</td>
<td>20</td>
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<tr>
<td></td>
<td>31-40 years</td>
<td>14</td>
<td>16</td>
</tr>
<tr>
<td></td>
<td>41-50 years</td>
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</tr>
<tr>
<td></td>
<td>&gt;50 years</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>3.</td>
<td>Domicile</td>
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<td></td>
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<tr>
<td></td>
<td>Rural</td>
<td>19</td>
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</tr>
<tr>
<td></td>
<td>Urban</td>
<td>14</td>
<td>11</td>
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<tr>
<td>4.</td>
<td>Educational</td>
<td></td>
<td></td>
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<td></td>
<td>Status</td>
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<tr>
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<td>Illiterates</td>
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<td>27</td>
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<td>7</td>
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<tr>
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<td>Secondary</td>
<td>19</td>
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<td>Collegeate</td>
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<td>5.</td>
<td>Occupation</td>
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<td>Housewives</td>
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<tr>
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<td>Salesman</td>
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<td>0</td>
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<td>Miscellaneous</td>
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2. SEROLOGICAL PROFILE OF HSV2 ANTIBODIES IN 16 HIV / AIDS PATIENTS

<table>
<thead>
<tr>
<th>S.No</th>
<th>HSVSero Positives</th>
<th>HIV Seropositives</th>
<th>AIDS</th>
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<tbody>
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<td>Male Adults</td>
<td>Female Adults</td>
<td>Male Adults</td>
</tr>
<tr>
<td>1.</td>
<td>IgM</td>
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<td>5</td>
</tr>
<tr>
<td>2.</td>
<td>IgG</td>
<td>4</td>
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3. SYMPTOMATOLOGY OF HSV2 ANTIBODIES POSITIVITY IN 16 HIV / AIDS PATIENTS

<table>
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<tr>
<th>S.No</th>
<th>Gender</th>
<th>Asymptomatic</th>
<th>Symptomatic</th>
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</thead>
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<td></td>
<td>IgM</td>
<td>IgG</td>
</tr>
<tr>
<td>1.</td>
<td>Males</td>
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<td>3</td>
</tr>
<tr>
<td>2.</td>
<td>Females</td>
<td>5</td>
<td>4</td>
</tr>
</tbody>
</table>

OBSERVATIONS

EPIDEMIOLOGICAL PROFILE:

Thirty-three male adults, 46 female adults and one female child HIV / AIDS patients were the material for this study. One male and 5 female adults were positive for HSV2 IgM antibody positives and four male adults and 6 female adults were positive for HSV2 IgG antibody.

Age profile showed that only one female child was below ten years old. Two females were in between 11 and 20, 8 males and 20 females, were in between 21 and 30 years. Fourteen males and 16 females were in between 31 and 40 years old. Eight males and 8 females were in between 41 and 50 years old. Only three males were above the age 50.

Regarding the domiciliary pattern, 19 males and 36 females were rural dwellers, Fourteen males and 11 females were urbanites.

Regarding the marital status, 27 males and 29 females were married. Two males and one female were unmarried. Four females were separated from the wedlock. Four widowers and 13 widows were reported in this study.
Regarding the educational status, 5 males and 27 females were illiterates. Nine males and 7 females were up to primary level of education. Nineteen males and 12 females were up to secondary education. Only one colleague were reported in this study.

Occupational profile showed 22 agricultural workers, 16 housewives, 8 drivers, 6 weavers, 4 sales men, 2 carpenters, 3 mechanics, 2 construction workers and 17 miscellaneous workers.

**CLINICAL PROFILE**

Thirty - two males and 45 females and one female child were HIV seropositives. One male adult and one female adult patients were in AIDS stage.

Pelvic inflammatory disease was diagnosed (cervical discharge, LBA, lower abdominal pain, irregular periods, menorrhagia and positive Chandler’s sign) in 23 adult females. Genital candidiasis was reported in 13 female adults. Trichomoniasis was reported in 13 female adults.

One male adult and one female adult had molluscum contagiosum. Only one female adult had nongonococcal Urethritis. Two males and three females were latent syphilits.

**Serological Profile**

Four male adults and five female adults HIV seropositive showed HSV IgG positivity. One male adult, five female adults and one female adult AIDS patient showed IgM antibody. Twelve HSV2 antibody positives (75 per cent) were asymptomatic during screening. Two patients with HSV2 IgG antibody positives (12.5 per cent) and two patients with HSV2 IgM antibody positives were symptomatics. One male adult and one female adult HIV seropositives with HSV2 IgG showed symptoms pertained HSV2.

One male adult HIV seropositive and one adult female AIDS patient with IgM antibody showed symptoms pertain HSV2.

**CD4 PROFILE**

Among HSV2 seropositives, only one AIDS patient’s CD4 T cell count was 196 cells/ cmm. Remaining 15 had CD4 counts between 320 cells/cmm to 420 cells/cmm.

**Endoscopic Study**

One AIDS female adult showed herpetic ulcers in the antrum of the stomach.

**HISTOPATHOLOGICAL PROFILE**

From Endoscopic biopsy in the antrum of the stomach showed nonspecific findings, but showed giant epithelial cells on smear study.

**DISCUSSIONS**

Herpes simplex virus type II seropositivity is usually associated with HIV acquisition and HIV infection exacerbates HSV2 infection. Persons with subclinical or unrecognized infection are best diagnosed by accurate type specific antibody tests. Paved the way to screen the HIV/AIDS patients for the same.

In our Study out of 80 HIV / AIDS patients screened, only in 16 patients, we have found out the HSV II seropositivity (20 per cent).

In our study, 5 males and 11 females were identified as HSV2 antibody positives. Too Small quantum to predict the sex preponderance.

Our study showed 15 HSV2 seropositives were in between 20 and 50 years old (sexually active period) simulated the literature.

Fourteen HSV2 seropositives were rural dwellers, indicated that the prevalence of this co-infection in rural community. Again too small quantum to predict the domiciliary pattern.

Regarding the marital status, all HSV2 seropositives in our study were married. The chance of infection transmission to the marital partner and to their offspring might be possible.

In 80 per cent to 100 per cent of adults, the increased frequency of HSV antibodies in lower socio-economic population appears to be due to a greater risk of becoming infection with HIV 1 and HSV 2. Nine HSV2 seropositives in our study were illiterates and two were up to primary education indicated the low literacy. Low literacy, ignorance, low socio-economic groups, lack of awareness, lack of understanding about the disease profile and spread appeared to be greater risk factor for acquisition of HIV 1 and HSV 2.

Our study showed that 10 HSV2 positives were promiscuous which may be the route cause for acquiring both HIV and HSV2.
In our study, 15 HSV2 seropositives were HIV seropositives and one female AIDS case showed HSV2 seropositivity. Most persons who have serologic evidence of infection with herpes simplex virus type 2 are asymptomatic. In our study, 12 (75 per cent) HSV2 antibody positives were asymptomatic during the screening.

Prevalence of symptomatic genito ulcerative disease (GUD) is increased both during the period of HIV seroconversion and after HIV acquisition. This increase is mainly observed among HSV2 seropositive individuals. In our study also, prevalence of 12.5 per cent IgG antibody positives and IgM antibody positives were recorded in symptomatic HSV patients.

In many persons, the first recognized reactivation may occur years after initial acquisition of HSV infection. In our study, 16 HSV2 antibody positive cases, we could not pinpoint the time of HSV2 infectivity occurrence prior or after HIV infection and about the reactivations, since due to the ignorance, poor observation, and understanding capacity.

Micro ulceration may have been exacerbated resulting in an increased frequency of symptomatic genito ulcerative disease after HIV acquisition, which may be considered as one of the factors in our symptomatic patients.

Heng et al noted that unique morphological and replicative characteristics of the virus in the co-infected cells which suggested the presence of HSV1/HIV1 hybrid virus in their study. It may be possible with HSV2 also. For want of facility, we could not detect this hybrid in our study. Thorough study of this co-infection may unfold the clinical variations and its therapeutic efficacy in HSV2/HIV1 hybrids if it exists. In future, this knot may be unfolded.

HIV & HSV2 co-infected individuals and their partners treatment of HSV2 infection may provide potential benefits against active genitoulcercative disease and that for HSV2 seropositive asymptomatic individuals, this treatment may provide prophylaxis. Regular followup may answer for this statement in future.

Type specific serological tests are useful in confirming a diagnosis of genital herpes and to diagnose persons with unrecognized infection and to manage sex partners of persons with genital herpes which may be true and useful in confirming the diagnosis of HSV2 in our patients.

In our study, one male and 5 females were HSV2 IgM positives may indicate the recent or current infection of HSV2.

For want of facility, we were not in a position to diagnose HSV2 by viral culture, Latex agglutination test, capture B/SA ELISA for detection of HSV antigen, Indirect haemagglutinatin test, complement fixation test, Immunoperoxidase assay, electron microscopic study for viral particles, immunofluorescence test, RIPA and PCR in this study.

With the increasing numbers of adult genital HSV infections and consequently increased neonatal HSV infection transmitted from vaginal deliveries, the rapid and accurate diagnosis of genital HSV infection are by the type specific serological tests for HSV2 antibodies.

Further a large-scale study with all investigative procedures may be helpful for accurate diagnosis in the near future and to study the clinical variations, exacerbations, and its significant effects one over the other virus.

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3. Our thanks are due to the Ethics Committee for giving permission to conduct this study.
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5. Our thanks are due to the patients who had co-operated in this study.
REFERENCES


