

Seroprevalence of HSV-2 in STI Clinic Attendees and Non High Risk Controls in a Tertiary Care Hospital in North India

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ABSTRACT

Introduction: Herpes Simplex Virus-2 is one of the major cause of Sexually Transmitted Infections leading to genital ulcer disease. Thus, an understanding of the pattern of STIs prevailing in different geographic regions of a country is necessary for implementation of STI control strategies. Therefore, seroprevalence of HSV-2 IgM and IgG in both cases and control group was studied along with the sociodemographic characteristics and proportion of primary and recurrent genital herpes.

Material and Methods: A prospective study was conducted in Department of Microbiology, G.M.C., Amritsar on blood samples obtained from 250 patients attending STI clinic and on 50 control group subjects attending Department of Skin and STD from Dec2015 to June 2017. Serological testing by ELISA for HSV-2 IgM and IgG antibody against glycoprotein G2 was performed.

Results: out of 250 cases, seroprevalence of HSV-2 IgM and IgG was 8.4% and 14% respectively. 35.7% cases of primary genital herpes and 64.3% recurrent genital herpes cases were observed. Male preponderance was noticed with maximum number of cases in the age group of 40-49years. Out of 50 controls, seroprevalence of HSV-2 was 8% ($p < 0.05$).

Conclusion: Due to the asymptomatic nature of HSV-2 and continuous shedding of the virus in majority of cases, there is a strong need to screen the vulnerable population like STI clinic attendees and other high risk groups for HSV-2 to protect the patients themselves and non-infected partners from acquiring these infections.

Keywords: Sexually Transmitted Infections, Herpes Simplex Virus-2, Genital Ulcer Disease

human papilloma virus infection were extremely rare. The increase in viral STIs may be attributed to greater self-reporting by patients, indiscriminate use of broad-spectrum antibiotics, effectiveness of syndromic approach of treatment and up-gradation of health services at the primary level.³ The spread of HIV since 1980s has also resulted in significant alteration in epidemiological patterns of STIs.

Genital herpes simplex virus (HSV) infection is the most common cause of genital ulcer disease globally.⁴ HSV has been characterized into two distinct serotypes; HSV-1 and HSV-2. Glycoprotein G2 (gG2) is a highly specific surface protein for detection of antibodies against HSV-2⁵ as known cross reactions do not occur with HSV-1. HSV-1 is more commonly associated with oro-pharyngeal disease while HSV-2 is more frequently associated with genital disease and is a good marker of high risk sexual behaviour in population. In Industrialized nations, HSV-2 is affecting more than one in five sexually active adults of 15-49 years age group with a global estimate of 536 million infected persons and an annual incidence of 23.6 million cases.⁶ The clinical course of first episode of genital herpes among patients with HSV-1 and HSV-2 infections is similar but the frequency and severity of recurrence is less with HSV-1 than with HSV-2. The IgM antibody is usually detectable after 9-10 days of primary infection. The time required for development of IgG antibody following HSV-2 infection varies from 21 days to over 42 days which is indicative of previous exposure. A significant increase in IgG titre in paired sera taken at an interval of 2 weeks is suggestive of reactivation of previous infection.⁷

Despite increased awareness of these infections, they remain under diagnosed because the majority of infections are asymptomatic or unrecognized.⁸ Diagnosis of genital herpes based on pure clinical grounds has been shown to have a poor sensitivity, so laboratory tests for confirmation need to be employed.⁷

INTRODUCTION

Sexually Transmitted Infections (STIs) are responsible for an enormous burden of morbidity, mortality and stigma in both developed and developing countries because of their effect on reproductive and child health.¹

Sexually Transmitted Diseases are transmitted from one person to another through sexual contact caused by a wide range of bacterial, viral, protozoal, fungal pathogens and ectoparasites resulting in clinical disease e.g. syphilis, genital herpes, chancroid, gonorrhoea, genital warts, Chlamydia and non-gonococcal urethritis. On the other hand, STIs are also transmitted through sexual interaction but do not cause clinical symptoms e.g. Hepatitis B, Hepatitis C, HIV and Human T Lymphotropic Virus-1 (HTLV-1). Now a days, the term STIs is commonly used rather than STDs.²

In India, during the 1960s and 70s, the major STIs were bacterial while viral diseases such as herpes simplex and

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Various laboratory methods for diagnosing genital herpes include direct microscopy, antigen detection, viral culture, polymerase chain reaction (PCR), and conventional type specific serology (TSS). Even though PCR is available but still TSS can be used as an effective tool in screening HSV-2 infection before initiating treatment since antibody detection is much easier, convenient, inexpensive and feasible screening test as compared to PCR and tissue culture.

Keeping these things in mind the study was conducted with the following aims and objectives:-

1. To estimate the seroprevalence of HSV-2 IgM and IgG antibody against glycoprotein G2 in both STI clinic attendees and control group.
2. Its correlation with socio-demographic characteristics

3. Assessment of the proportion of primary and reactivated HSV-2 cases.

MATERIAL AND METHODS

This observational descriptive study was undertaken in the Department of Microbiology, Government Medical College, Amritsar from December 2015 to June 2017. 250 patients were enrolled belonging to 18-49 years of age, attending STI clinic in the Department of Dermatology, Government Medical College, Amritsar with clinical features suggestive of STIs and patients with previous history of treatment now presenting with suggestive clinical features. 50 subjects attending the Dermatology OPD for complaints other than STIs were enrolled in Control Group. Blood samples

	HSV-2 IgM seropositive	%	HSV-2 IgG seropositive	%	Both HSV-2 IgM and IgG seropositive	%	P value
Cases (n=250)	21	8.4	35	14	3	1.2	0.013 (significant)
Controls (n=50)	-	-	4	8	-	-	

Table-1: Serological profile of HSV-2 IgM and IgG in cases and controls

Herpes genitalis	Number of cases (n=56)	%	HSV-2 IgM sero positive	%	HSV-2 IgG seropositive	%	Both HSV-2 IgM and IgG sero-positive	%	HSV-2 seronegative	%
Primary	20	35.70	6	30	3	15	1	5	10	50
Recurrent	36	64.30	-	0	14	38.9	2	5.5	20	55.6

Table-2: Distribution of herpes genitalis cases

Variables	Cases (n=250)	Percentage (%)	HSV-2 seropositive cases (n=59)	Percentage (%)
Age (years)				
18-28	80	32	15	25.4
29-39	65	26	16	27.1
40-49	105	42	28	47.5
Sex				
Male	168	67.2	44	74.6
Female	82	32.8	15	25.4
Education (p value-0.008 significant)				
Illiterate	27	10.8	7	11.9
Upto matric	168	67.2	42	71.1
Above matric	55	22	10	17
Number of sexual partners				
Multiple	81	32.4	36	61
Single	169	67.6	23	39
Type of sexuality				
Heterosexual	248	99.2	59	100
Homosexual	2	0.8	0	0
Condom usage				
No	42	16.8	5	8.6
Occasional	180	72	53	89.8
Yes	28	11.2	1	1.6
Age at first sexual contact (years)				
15-25	233	93.2	54	91.5
26-36	14	5.6	5	8.5
36-48	3	1.2	0	0

Table-3: Distribution of cases according to socio-demographic characteristics and risk factors

Provisional Diagnosis	Number of Cases (n=250)	%	HSV-2 seropositive cases (n=59)	%
Anal ulcer	3	1.20	1	1.8
Anal warts	4	1.60	1	1.8
Balanitis	46	18.40	5	8.5
Balanoposthitis	54	21.60	9	15.2
Genital warts	24	9.60	6	10.1
Herpes genitalis	56	22.40	26	44
Molluscum contagiosum	14	5.60	6	10.1
Vaginal candidiasis	24	9.60	3	5.1
Vulvovaginitis	25	10.00	2	3.4
Total	250	100.00	59	100.00

Table-4: Provisional diagnosis of cases (n=250)

were collected after taking written informed consent along with the detailed history taken on predesigned proforma. The observations were based on serological testing by Euroimmun ELISA test kit for HSV-2 IgM and IgG against Glycoprotein G2. The results were tabulated and analyzed by utilizing Chi-square test.

RESULTS

The serological profile of HSV-2 IgM and IgG in cases and controls both have been shown in Table-1. The correlation of HSV-2 seropositive cases with various socio-demographic characteristics and risk factors has been denoted in Table-2. The distribution of cases according to provisional diagnosis has been depicted in Table-3. The distribution of primary and recurrent herpes genitalis cases along with their serological profile is depicted in Table-4.

DISCUSSION

Sexually transmitted infections (STIs) constitute a major public health problem for both developed and developing countries. During the past two decades STIs have undergone a dramatic transformation. Presently, viral STIs like Herpes genitalis are emerging as most common cause of STI. Many people with common STDs remain asymptomatic and without diagnosis or even after diagnosis do not continue treatment and serve as reservoir of infection and thus transmit infection to their sexual partners.⁹ There is a dearth of information regarding the epidemiology of STIs in India for reasons such as stigma and discrimination associated with them resulting in poor attendance of patients at health care centres.¹⁰ Thus, a proper understanding of the pattern of STIs prevailing in different geographic regions of a country is necessary for proper planning and implementation of STI control strategies.² The rationale for TSS is to accurately differentiate antibody response generated by HSV-1 from HSV-2 with the help of glycoprotein G (gG) and identify asymptomatic HSV infection. TSS as compared to PCR is easier, inexpensive, convenient and feasible screening test which can also detect silent carriers of HSV-2 infection especially in high risk settings such as STIs.¹¹ If genital lesions are present, type-specific serology can aid in establishing the diagnosis of a new episode or reactivation of HSV infection. The distinction between newly acquired HSV and reactivated HSV is helpful for epidemiological

studies.¹²

In present study, statistically significant association (p value=0.013) was found between the seroprevalence of HSV-2 in STI clinic attendees and control group accounting for 23.6% and 8% respectively (Table-1). Out of 50 control group subjects none of them was found to be positive for HSV-2 IgM. Chawla et al¹³ also noticed comparable results. Our study showed that 59 (23.6%) STI clinic attendees (study group) were HSV-2 seropositive of which 21 (%) were seropositive for HSV-2 IgM and 35 (%) for IgG (Table-2). We also observed that 3 cases were seropositive for both HSV-2 IgM and IgG which was comparable to the results noticed by Amudha et al⁶ and Choudhry et al¹⁰ with 3 (0.0329%) and 2 (0.02%) cases respectively. The provisional diagnosis of primary and recurrent genital herpes was observed in 20 (35.7%) and 36 (64.3%) cases respectively. Out of 20 primary genital herpes cases 6 (30%), 3 (15%) and 1 (5%) cases were found to be positive for HSV-2 IgM, HSV-2 IgG and both HSV-2 IgM and IgG respectively. Out of 36 recurrent genital herpes cases 14 (38.9%) were found to be positive for HSV-2 IgG and 2 (5.5%) cases were seropositive for both HSV-2 IgM and IgG. The presence of HSV-2 IgG antibodies in patients of primary herpes genitalis can be explained by the fact that HSV-2 IgM antibodies may take up to 10 days from exposure to develop and lasts only for 7–10 days. After that, seroconversion to IgG antibodies takes place which can be detected if patient reports late for treatment.⁶ The presence of HSV-2 IgM antibodies in recurrent genital herpes cases can be related to the fact that IgM appears sporadically in about one third of patients after recurrent genital herpes episodes.¹⁴

Our study showed that maximum number of cases i.e. 105 (42%) belong to the age group of 40-49years with the mean age of 35.21 \pm 9.76 years (Table-3) which is comparable to the study conducted by Patwardhan et al¹⁵ and Sethia et al¹⁶ with predominant age group being 36-45 years and >35 years respectively. Also maximum seropositivity of HSV-2 in our study was seen in the age group of 40-49years i.e. 28 (47.50%) cases which was comparable to the study conducted by Amudha et al.⁶ Older people may be affected by STIs because they are more often diagnosed with recurrent diseases rather than primary infections. Moreover, as age increases, the number of years of sexual activity also

increases which in turn leads to repeated exposures to high risk sexual behavior and contracting various STIs during their lifetime.

In our study, male preponderance was seen in 168 (67.20%) cases (Table-3) which is in concordance with the study conducted by Jain et al¹⁷ and Devi SA et al.¹⁸ We also observed that seroprevalence of HSV-2 was more in males accounting for 44 (74.60%) cases. This increased seropositivity amongst males was observed due to the promiscuity of men along with significant change in sexual behaviour due to premarital and extramarital sexual contact.

The educational status analysis revealed that majority of the cases i.e. 168 (67.2%) were educated up to matric which was found to be statistically significant (p value=0.008) (Table-1). We also observed that 27 (10.80%) cases were illiterate and 55 (22%) cases were educated above matric. Similar findings were observed in a study conducted by Nair et al.¹⁹ Our study also showed that majority of seropositive HSV-2 cases were educated upto matric accounting for 42 (71.1%) cases which is consistent with the study conducted by Amudha et al.⁶ This shows the importance of literacy level as it leads to better understanding and awareness which in turn leads to lesser incidence of STIs amongst educated people.

Various risk factors associated with STIs are multiple sexual partners, condom usage, age at first sexual contact and type of sexuality. Out of all these risk factors studied, most common association of STI was found to be with the H/o multiple sexual partners i.e. 81 (32.4%) which was statistically significant also (p value=0.000). Comparable results were seen in a study conducted by Nayyar et al.²⁰ Contact with multiple sexual partners was found to be the major risk factor amongst the HSV-2 seropositive cases accounting for 36 (61%) cases. In our study we observed that majority i.e. 248 (99.20%) cases were heterosexual while only 2 (0.80%) cases were homosexual (Table-3) which was in concordance with the findings of studies conducted by Devi SA et al.¹⁸ This shows that sexual behavior is critical for understanding the spread of STIs.

In our study, only 28 (11.20%) cases reported regular condom usage (Table-3). Comparable results were seen in the studies conducted by Devi SA et al¹⁸ and Nayyar et al.²⁰ We also observed that 180 (72%) cases admitted of using condom inconsistently while 42 (16.80%) cases never used barrier contraceptives which is in concordance to the study conducted by Nayyar et al.²⁰ In our study majority of the seropositive cases of HSV-2 i.e. 53 (89.8%) used condom inconsistently. This may be attributed to the fact that the individuals who do not use condom or use it inconsistently are more prone to STIs.

In the present study, 233 (93.20%) cases reported their first sexual contact in the age group of 15-25 years. These findings were similar to the study conducted by Nair et al¹⁹ and Choudhry et al.¹⁰ We also observed that maximum number of seropositive cases of HSV-2 i.e. 54 (91.5%) had their first sexual contact in the age group of 15-25 years. This may be attributed to the curiosity amongst teenagers regarding sexual activity, thus making persons vulnerable for

high risk sexual practices.

Herpes genitalis was the main provisional diagnosis in maximum number of cases i.e. 56 (22.40%) followed by 54 (21.6%) and 46 (18.4%) cases of Balanoposthitis and Balanitis respectively. Comparable results were reported by Jain et al¹⁷ and Devi et al.¹⁸ The provisional diagnosis of Herpes genitalis predominated in HSV-2 seropositive cases comprising of 26 (44%) individuals. 33 (56%) HSV-2 seropositive cases did not give H/o any symptoms suggestive of Herpes genitalis. This is because most of the individuals infected with HSV are asymptomatic or have very mild symptoms which remain unnoticed or may be mistaken for another skin condition. Shivaswamy et al²¹ and Varela et al²² also observed similar findings.

CONCLUSION

Westernization of culture, migration for jobs, weakening of traditional familial and social control on sexual behavior and decline in joint family has resulted in increasing burden of STIs in our society. However, syndromic management approach has been a major step in rationalizing and improving the treatment of STIs though with some shortcomings. More recently, viral STIs like genital herpes are emerging as predominant STI. Due to the asymptomatic nature of HSV-2 and continuous shedding of the virus in majority of cases, there is a strong need to screen the vulnerable population like STI clinic attendees and other high risk groups for HSV-2 where Type specific serology (TSS) plays an important role to protect the patients themselves and non-infected partners from acquiring these infections. Awareness programs should include sexual education, counseling and voluntary testing targeted to all sexually active age groups especially high risk groups regarding safe sexual practices, various STIs and its complications.

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