SEROPREVALENCE OF HERPES SIMPLEX VIRUS 1 & 2, HEPATITIS B AND CHLAMYDIA TRACHOMATIS AMONG THE PATIENTS ATTENDING SEXUALLY TRANSMITTED DISEASES DEPARTMENT

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CERTIFICATE

Certified that this dissertation entitled **"SEROPREVALENCE OF HERPES SIMPLEX VIRUS 1 & 2, HEPATITIS B AND CHLAMYDIA TRACHOMATIS AMONG THE PATIENTS ATTENDING SEXUALLY TRANSMITTED DISEASES DEPARTMENT"** is a bonafide work done by **DR. G. SUBHA**, Post Graduate Student in **M.D. Dermatology, Venereology and Leprosy**, Madras Medical College, Chennai-600 003, during the academic year 2007-2010.This work has not previously formed the basis for the award of any degree.

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CONTENTS

INTRODUCTION

Sexually transmitted diseases (STDs) are a major cause of morbidity in developed as well as developing countries. STD incidence rates remain high in most of the world, despite diagnostic and therapeutic advances that can rapidly render patients with many STDs noninfectious and cure most.

In 1996, the World Health Organization estimated that more than 1 million people were being infected daily. About 60% of these infections occur in young people <25 years of age, and of these 30% are <20 years. Between the ages of 14 and 19, STDs occur more frequently in girls than boys by a ratio of nearly 2:1; this equalizes by age 20. An estimated 340 million new cases of syphilis, gonorrhoea, chlamydia and trichomoniasis occurred throughout the world in 1999.

Centre for Diseases Control and Prevention (CDC) has estimated that nearly 20 million STD cases occur every year, with half among people less than 25 years.

Changing cultural values, changing sexual morals, increase in travel, all had contributed for increase in STDs. Additionally,

development and spread of drug resistant bacteria (e.g., penicillin-resistant gonococci) makes some STDs harder to cure.

Commonly reported prevalence of Sexually Transmitted Infections (STI) among sexually active adolescent girls both with and without lower genital tract symptoms include Chlamydia (10–25%), gonorrhoea (3–18%), syphilis (0–3%), Trichomonas vaginalis (8–16%), and herpes simplex virus infection (2–12%). Among adolescent boys with no symptoms of urethritis, isolation rates include chlamydia (9–11%) and gonorrhea (2–3%).

HERPES SIMPLEX VIRUS INFECTIONS

Herpes simplex virus is a DNA virus belonging to Herpesviridae family.

The word Herpes means ‘to creep’. Latency is a common property of the entire herpes virus.

EPIDEMIOLOGY

Genital herpes infection assumes great importance in the HIV era. HSV2 infections are acquired between 15 and 40 years of age. Most of the genital herpes are caused by HSV2; however the incidence of genital herpes caused by HSV1 is increasing. Men are more likely to acquire

HSV2 asymptomatically than women. Asymptomatic viral shedding is the primary mode of herpes virus transmission.

PATHOGENESIS

A viral infection with HSV induces ballooning of cells, followed by degeneration of nucleus and later multinucleate giant cells are formed. The infected person can excrete the virus from peripheral sites, mucosal, genital or oral secretions.

The virus enters the abraded site, replicates, followed by retrograde transport to dorsal nerve root ganglion (trigeminal ganglion in HSV1 and sacral ganglion in HSV2). The prior immune response to HSV1 has protective effect on the acquisition of HSV2.

CLINICAL MANIFESTATIONS

HSV1 infections range from asymptomatic infection to symptomatic infections in the form of fever, sore throat, ulcerative and vesicular lesions, gingivostomatitis, oedema and localized lymphadenopathy. The incubation period ranges from 2 – 12 days. HSV1 is also known to cause primary genital herpes.

PRIMARY GENITAL HERPES

Primary genital herpes can be caused by both HSV-1 and HSV-2 and can be asymptomatic. The clinical features and course of primary genital herpes caused by both HSV-1 and HSV-2 are indistinguishable, but recurrences are more common with HSV-2. It is manifested by prodromal symptoms like fever, headache, malaise, myalgia, followed by vesicles rupturing into ulcers. The lesions persist for 3 weeks without treatment.[1]

Women with primary infection manifest with lesions in vulva, the cervix being variably involved. They are more painful and associated with inguinal lymphadenopathy and dysuria.

Males present with vesicles over glans penis, penile shaft, inner aspect of prepuce, thighs, gluteal regions and perineum.

The complications following primary herpes infection is more such as urinary retention, neuralgia and meningoencephalitis.

The non primary genital herpes presents with less severe symptoms and heals quickly.

Recurrent herpes infections present with less number of lesions. The frequency of recurrences varies with individuals. Immunosuppressed

patients have frequent and prolonged reactivation of latent HSV infection. They present with atypical manifestations like vesicles which coalesce into deep necrotic ulcers and vegetative plaques.

Various studies have shown that prior HSV2 infection is associated with acquisition of HIV infection. Asymptomatic viral shedding occurs more with HIV positive persons.[2]

LABORATORY DIAGNOSIS

1) Microscopy (Tzanck smear)

Staining of the scrapings from the floor of the ulcer and roof of vesicle with Wright or Leishman stain shows the presence of multinucleate giant cells and epithelial cells containing eosinophilic intranuclear inclusion bodies. It cannot differentiate between HSV1 and HSV2 and the sensitivity is 67%.

2) Culture of HSV

Herpes simplex virus infection is best confirmed by isolation of the virus in tissue culture. Tissue culture success is operator-dependent, but this modality can yield positive results within 48 hours of inoculation.

Characteristic cytopathic effect with ballooning of cells and cell death are observed, and death of the entire monolayer of cells may be rapid.

Immunofluorescent staining of the tissue culture cells can be used to quickly identify HSV and can distinguish between types 1 and 2.

3) Detection of HSV antigen

    a) Immunofluorescence

    b) Immunoperoxidase

    c) Enzyme Immuno Assay

4) Detection of HSV DNA by PCR

5) Type specific serological assays

These assays had revolutionized the accurate evaluation of seroprevalance of HSV1 and HSV2, the advantage being detection of HSV2 antibodies in the presence of HSV1 antibodies and vice versa. The Enzyme Linked Immunosorbent Assay (ELISA) uses purified type specific proteins. The sensitivity is 85 – 98% and specificity is >95% based on the kit used.

HEPATITIS B INFECTION

Hepatitis B virus (HBV) infection can be either acute or chronic, ranging in severity from being asymptomatic to symptomatic and completely resolving to severe, progressive and even fatal illness.

EPIDEMIOLOGY

The carrier rate of Hepatitis B ranges from 0.1% to 20% in different parts of the world.

TRANSMISSION

Transmission of HBV is largely from a parenteral or in apparent parenteral route.

Parenteral transmission:-

HBV infection can occur from exposure to blood from chronic carriers of Hepatitis B virus. HBsAg is found in low concentrations in the urine, breast milk, vaginal secretions, cerebrospinal fluid, sweat, tears, bile and feces. Transmission thus occurs with blood transfusion, unpasteurized plasma products, needle stick injuries, and unsterile instruments when used in tattooing, acupuncture, ear piercing or dentistry.

Sexual transmission:-

It is the most important mode of HBV transmission.

Mostly occurs with receptive anal intercourse and homosexual population with multiple sexual partners. Silent carriers form an important source of HBV transmission.

In apparent transmission:-

It may occur in family through saliva, blood tinged fluid, fluid from open wounds and patch of dermatitis.

Perinatal transmission:-

The HBsAg positive mother is likely to transmit the infection to the neonate in 90% of cases.

PATHOGENESIS

Immunological factors play an important role in causing liver disease due to HBV. Acute hepatitis is due to immune lysis.

Chronic infection is more common in immunosuppressed patients, patients on hemodialysis, post renal transplant recipients, patients with leprosy or leukemia and those with associated HIV infections.

CLINICAL FEATURES

The spectrum of HBV infection during the acute phase varies from subclinical hepatitis, anicteric hepatitis, icteric hepatitis to fulminant hepatic failure.

Incubation period ranges from 40 to 140 days.

EXTRAHEPATIC MANIFESTATIONS OF HBV INFECTION

The extrahepatic manifestations include serum sickness, polyarthritis nodosa, membraneous glomerulonephritis, papular

acrodermatitis, aplastic anemia, myocarditis, pericarditis and neuromuscular complications. These are believed to be mediated by immune complexes in circulation.

CHRONIC HEPATITIS

The chronic phase ranges from an asymptomatic carrier state to chronic hepatitis, cirrhosis and ultimately hepatocellular carcinoma.

SEROLOGICAL DIAGNOSIS

- The first serological marker to be detectable in the serum is HBsAg. It appears during the incubation period.[3] HBsAg appears before the onset of symptoms, peaks during overt disease, and then declines to undetectable levels in 3-6

months. Acute HBV infection is characterized by the presence of HBsAg in the serum.

- HBeAg, HBV DNA, and DNA polymerase appear in the serum soon after HBsAg, and all signify active viral replication. Measuring HBV DNA with quantitative DNA polymerase chain reaction (PCR) is ideal for monitoring disease progression and effect of treatment.

- Immunoglobulin M (IgM) anti-HBc becomes detectable in serum shortly before the onset of symptoms, concurrent with the onset of

- elevation of serum aminotransferases. Over months, the IgM antibody is replaced by immunoglobulin G (IgG) anti-HBc .

- Anti-HBe is detectable shortly after the disappearance of HBe Ag, implying that the acute infection has peaked and the disease is on the wane.

- IgG anti-HBs does not rise until the acute disease is over and is usually not detectable for a few weeks to several months after the disappearance of HBsAg. Anti-HBs may persist for

life, conferring protection; this is the basis for current vaccination strategies using noninfectious HBsAg.

- During convalescence, HBsAg and HBe Ag are cleared, and IgG antibodies to HBsAg, HBcAg, and HBeAg develop.

- Hepatitis B surface antibody (HBsAb) is a protective antibody that neutralizes the virus, although the coexistence of HBs Ag and HBsAb has been reported in approximately 25% of individuals who are HBsAg positive. HBsAb, but not hepatitis B core antibody (HBcAb), is detected in persons who have received the hepatitis B vaccine.

- Total HBcAb, including IgM and IgG, indicates exposure to the virus and viral replication. HBcAb appears shortly after HBsAg in

- acute disease and persists for life; therefore, HBcAb is not a good marker for acute disease.

- Detection of IgM HBcAb is diagnostic of acute HBV infection.

- The carrier state is defined by the presence of HBsAg in serum for 6 months or longer after initial detection. The presence of HB sAg alone does not necessarily indicate replication of complete virions, and patients may be asymptomatic and without liver damage.

- In contrast, chronic replication of HBV virions is characterized by persistence of circulating HBsAg, HBeAg, and HBV DNA, usually with anti-HBc and, occasionally, with anti-HBs. In these patients, progressive liver damage may occur.

- The major clinical role of serum HBV DNA assays is the assessment of the candidacy of patients with chronic HBV infection for antiviral therapy and their response to it. Tests for HBV DNA in serum rarely help in identifying HBV as the cause of liver disease in patients who are HBsAg-negative; knowledge of this fact is especially important in patients with fulminant hepatitis B in whom HBsAg may have cleared by the time they seek.

CHLAMYDIA TRACHOMATIS

The chlamydiae are non motile, gram negative, obligate intracellular bacteria. Chlamydia trachomatis pathogens can be divided into at least 15 serovars. The A, B, Ba, C serovars are associated with hyperendemic blinding trachoma. The D – K serovars cause sexually transmitted disease. The L1, L2, and L3 serovars cause lymphogranuloma venereum.

EPIDEMIOLOGY

The chlamydial infections are much more widely dispersed. The risk factors include previous chlamydial infection, recent change in partner, asymptomatic partner, failure to use barrier contraceptives, low socioeconomic status [5,7,8] and gonorrhea infections.

CLINICAL FEATURES

The most common manifestation is non gonococcal uretheritis (NGU) in men. It is characterized by pyuria. NGU is diagnosed by demonstrating a significant number of polymorphonuclear cells in the first catch urine or a smear prepared from a urethral swab.

Postgonoccal uretheritis occurs in men who had been successfully treated for gonococcal uretheritis. It can cause epididymitis in sexually active men. Rectal infections are also common.

In females, the most commonly affected site is the cervix. The organism can cause mucopurulent endocervicitis. Chlamydia trachomatis is a parasite of columnar epithelium. It also causes sterile pyruia and ascending genital infection. Acute salphingitis is the most important complication of sexually transmitted chlamydial infection. [4,6,9] Chlamydia trachomatis is also associated with the Fitz-Hugh-Curtis syndrome.

The Chlamydia can also cause mucopurulent conjunctivitis and pneumonia in infants [10,11,12]

In lymphogranuloma venereum, the primary lesion is painless and often missed.[10] Secondary lesions are characterized by the involvement of lymph nodes. Both inguinal and femoral nodes can be involved. Later it may lead to suppuration and rupture of the nodes, developing draining fistulas. The rare manifestations of LGV include meningitis, arthritis, pneumonia and conjunctivitis.

LABORATORY DIAGNOSIS

Cytology

The infection can be diagnosed by demonstrating typical intracytoplasmic inclusions on cytological examination.

Fluorescent antibody technique

This is based on detecting elementary bodies in smears.

Enzyme Immunoassay

In this the chlamydial antigens are detected especially chlamydial lipopolysaccharide.

Culture

The most common technique involves inoculation of specimens into cycloheximide treated McCoy or other appropriate cell lines. Intracytoplasmic inclusions can be detected either by Giemsa stains or by immunofluorescent staining with monoclonal antibodies. This has high specificity (100%) and sensitivity.

Nucleic Acid Amplification Tests

- Polymerase Chain Reaction (PCR),
- Ligase chain reaction,

- Transcription mediated amplification
- Strand displacement assay

Serological tests

They are less useful in genital infections but play a supportive role in establishing diagnosis in chlamydial salphingitis or epididymitis. The Micro – IF test is the serological test of choice.[13]

REVIEW OF LITERATURE

Gopi Thawani et al [14] from Calcutta performed a serological study for sexually transmitted diseases in patients attending STD clinics in Calcutta. Out of 457 samples studied positivity was highest with TPHA (18.60%), followed by chlamydia (15.97%), VDRL (8.98%), HIV (6.35%) and HBsAg (3.72%). A total of 37.20% samples were positive. Maximum infection was seen in 15-30 years age group (20.13%), followed by 30-45 years age group (12.69%) and > 45 years age group (4.38%). Single test, two tests and four tests positivity was highest in 15-30 years age group but three tests positivity was highest in 30-45 years age group. Positivity was maximum in single test (65.29%) followed by two tests (25.88%), three test (8.24%) and four tests (0.59%).

Amongst the single test positive, maximum was with chlamydia (36.94%), followed by TPHA (30.63%) HIV (18.91 %), VDRL (7.21%) and HBs Ag (6.31%). Amongst two test positivity VDRL + TPHA positivity was the highest (45.45%). Other combinations were TPHA+ chlamydia (29.54%), chlamydia + HIV (9.09%), HBsAg + Chlamydia (4.55%), TPHA + HIV (4.55%), HBsAg +HIV (4.55%) and TPHA + HBsAg (2.27%). Amongst the positivity in three tests, VDRL+ TPHA + Chlamydia positivity was maximum (71.43%). Other combinations were TPHA + chalmydia + HBsAg (14.28%) and VDRL + TPHA +HBsAg

(14.28%). Only one sample was positive in 4 tests ie VDRL + TPHA +HBsAg+ chlamydia and it was 15-30 years age group. Out of 457 samples tested, 422 were males and 35 females. Positivity was also more in males (31.73%) than in females (5.47%).

A study by Jindal et al [15] from Amristar in the year 2007 studied the prevalence of HIV, HBV and HCV among 350 STD clinic attendees. The prevalence was 4.3% for HIV and 3.7% for HBV. Out of the 350 patients, 2.0% had co infection of HIV and HBV.

Rishbud et al [16] in the year 2002 studied the prevalence and incidence of HBV among patients attending three STD clinics in Pune. Of the 497 participants 3.6%, 26.5%, and 43.2% were positive for

HBsAg, anti-HBs, and anti-HBc respectively. 72 out of 497 (14.5%) participants were HIV positive. Tattooing was found to be independently associated with presence of core antibody. Additionally, history of being in commercial sex work and history of a genital ulcer were independently associated with a positive anti-HBc antibody test.

Rajesh et al[17] from Perundurai, Tamilnadu studied the seroprevalence of HBV and HCV in patients attending STD clinic. Out of 105 patients screened 5.71% were HBV positive and 1.90% for HCV. Male to female ratio was 6.6:1. Most were in the age group of 30 – 34

years followed by 25 – 29 years. None of their patients were symptomatic for the disease. HIV and HBV co infection was seen in 2.85% patients. Also another 2.85% patients had co infection of HIV and genital herpes.

Singh S et al[18] from Puducherry studied the seroprevalence and risk factors for transmission of HIV, HBV and HCV using anti – HIV, HBsAg and anti – HCV markers in serum. The positivity rates of HIV, HBsAg and HCV were 23.20 per cent, 10.00 per cent and 21.10 per cent respectively. The HIV and HBsAg positivity was higher in the reproductive age group while HCV positivity was higher in the elderly individuals (>40 years). Higher positivity rates of all three infections

were found in females, patients engaged as drivers, bisexuals, patients with an HIV infected spouse or contact with paid partners (CSW's).

Chopra A et al[19] from Amristar studied the HBsAg seroprevalence in Patiala. Out of 50 patients enrolled, 70% were males and 30% were females. 80% of the patients were in the sexually active age group of 15-34 years. HBsAg positivity was found in 10% of cases. Out of these positive cases 80% were males and 20% were females and 80% of these cases were in the age group of 25-34 years while only 20% were in between the age of 35-44 years. None of these cases were below 25 years or above 45 years of age. 60% of these cases had sex with more than 8

partners while 20% of each had 5-8 and 1-4 partners. VDRL test was positive in 38% of the cases. In VDRL positive cases 10.52% were HBsAg positive. Out of the remaining VDRL negative cases only 3.22% were positive for HBsAg.

Ray et al[20] from New Delhi analysed their STD clinic statistics for a period of 15 years from 1990 to 2004. In this retrospective study the prevalence of HIV was 2.2% for STD clinic attendees and 4.3% for STI cases. The seropositivity for HSV2 IgM among patients presenting with genital ulcers was 22.4%.

Aggarwal et al[21] from Amristar studied the seroprevalence of HSV1 and HSV2 antibodies among STD clinic attendees. The study was conducted on 250 serum samples of STD clinic patients and 50 serum samples of asymptomatic women to determine seroprevalence of herpes simplex-1 and 2 (HSV-1 and 2) IgM antibodies and HIV-1 and 2 antibodies. The samples were also screened for syphilis by VDRL test and confirmed by TPHA test. Seropositivity of HSV in STD clinic patients was 44/250 (17.6%) and 12/50 (24%) in asymptomatic women. In 11/44 (25%) seropositive persons for HSV, HIV 1 and 2 antibodies were present. In 10/44 (22.7%) HSV seropositive persons, coinfection with syphilis was also present, whereas in 7/44 (15.9%) HSV seropositive

persons, both HIV and syphilis were present. In the control group, co infection with other sexually transmitted infections (STIs) was not observed.

In a case control study done by KN shivasamy et al[22] from JIPMER, Puducherry, they analysed 135 patients who attended STD clinic, along with 135 age and sex matched controls. Among study group cases, 112 (82.9%) cases were co-infected with HSV-1 and HSV-2, 11 (8.1%) cases were seropositive for HSV-1 alone and 3 (2.2%) cases were

seropositive for HSV-2 alone. In the control group, 112 (82.9%) cases were co-infected with HSV-1 and 2, 12 (9.6%) for HSV-1 alone and 1(0.8%) for HSV-2 alone. Amongst the 53 cases (73.6%) of ulcerative STDs, 37 cases (63%) were clinically diagnosed as having genital herpes, of which 9 cases (24.3%) were of first episode genital herpes and 28 cases (75.6%) were of recurrent genital herpes. In the first episode genital herpes group, 6 cases (66.6%) each were positive for HSV-1 and HSV-2, and 5 cases (55.5%) were co-infected with HSV-1 and 2. In the recurrent genital herpes group, 27 cases (96.4%) each were positive for HSV-1, HSV-2 and both HSV-1 and 2.

Barbara Suligoi et al[23] did a cross-sectional study to determine the seroprevalence and the risk factors for HSV-2 infection among 776 HIV-

negative persons attending an STD clinic in Milan, Italy. All samples were tested with a commercial HSV type-2 specific IgG ELISA test. The HSV-2 seroprevalence was 29.5%. The seroprevalence increased with age, yet it did not differ by gender. Among persons with a current STD, the seroprevalence was 44.3%. At the multivariate analysis, older age was independently associated with HSV-2 infection. A self-reported history of genital herpes was predictive of HSV-2 infection. They concluded that the agreement between history of genital herpes and

HSV-2 seroprevalence was poor; however, caution should be used in interpreting the presence or absence of a history of genital herpes as an indicator of the presence or absence of HSV-2 infection.

N Langeland et al[24] examined the patients attending a referral sexually transmissible diseases clinic at Muhimbili Medical Centre in Dar-es-Salaam, Tanzania during the period 1989 to 1993 for herpes simplex virus type 2 (HSV-2) antibodies. An ELISA technique, using glycoprotein G of HSV-2 as antigen, was used to test 294 patients' sera. Of these, 126 sera were HSV-2 positive, while 168 were negative, yielding an overall HSV-2 prevalence of 42.9%. Sixty-three per cent of the women and 35.5% of the men were HSV-2 positive. Seropositivity rose from 8.7% in the youngest men to 61.5% in the oldest male age group, while even the youngest women aged 20 or less had an HSV-2

prevalence of 55.6%. There was a significant positive association between HIV and HSV-2 seropositivity (P =0.0006), most pronounced among the youngest women.

Marco Cusini et al[25] from Milan, Italy did a cross-sectional study to ascertain the HSV-2 prevalence among 919 persons attending an STD clinic in northern Italy. A prevalence of 24.6% was found without differences between males and females. Seroprevalence increased with

age and number of partners during the previous year. Seroprevalence significantly increased with age, ranging from 14.2% among persons 25 years or younger to 37.5% among persons older than 35 years. There was no significant difference between the prevalence found among homosexual or bisexual males (22.3%) and heterosexual males (27.8%).

A large randomized controlled trial was conducted from July 1993 to September 1996 to determine the seroprevalence and correlates of herpes simplex virus type 2 infections (HSV-2) in a geographically dispersed population of US among patients attending five STD clinics under the name Project RESPECT[26]. Of 4,128 total participants, 2,348 (56.9 percent) were male and 1,780 (43.1 percent) were female. Overall, 1,686 (40.8 percent) of the 4,128 participants were positive for HSV-2 antibody. HSV-2 seroprevalence was higher among women (52 percent)

than among men (32.4 percent). Independent predictors of HSV-2 seropositivity included female sex, black race, older age, less education, more lifetime sex partners, prior diagnosis of syphilis or gonorrhoea, and lack of HSV-1 antibody. Four additional factors predicted HSV-2 infection in women: a prior diagnosis of trichomoniasis, a current diagnosis of trichomoniasis, history of prostitution, and having a sex partner who had been in jail. The only additional factor predicting HSV-2

infection in men was a history of being in jail. Circumcision was not independently associated with HSV-2 infection. The majority of HSV-2-seropositive persons (84.7 percent) had never received a diagnosis of genital herpes.

Choudhry S et al[27] from Delhi studied the serological profile of HSV2 serology among patients attending STD clinic in Delhi. The study enrolled 100 patients and 30 were diagnosed clinically as genital herpes. Out of those 30, 17 (56.6%) were positive for HSV2 IgM. 7 (23.33%) were seropositive even though the clinical features were not suggestive of genital herpes. 6 (20%) were seronegative even though clinically diagnosed as genital herpes. 8 patients had other concomitant diseases.

Mahendra M Kura et al[28] studied the pattern of sexually transmitted diseases (STD) among two hundred and fifteen consecutive first-time

STD clinic attendees at a teaching hospital in Bombay in October 1995. Ulcerative STD constituted 73.5% of total STD while 15.8% were genital discharges and 10.2% were genital growths. Ulcers in decreasing order of frequency were chancroid (51.9%), genital herpes (29.1%) and syphilis (14.5). 76.5% of genital discharges were due to gonococcal infection. Of 182 patients tested for HBV, 16 (8.8%) were reactive for HBsAg.

Joyee et al[29] studied the seroprevalence of Chlamydia antibodies in serum and their correlation with PCR for Chlamydial antigens in genital swab in the year 1998 to 2002 in Government General Hospital , Chennai. Out of 143 symptomatic patients studied serologic positivity by IgM, IgA and IgG was 22.4%, 28.7% and 58.7% respectively. The PCR analysis showed 44 (30.8%) cases with confirmed C. trachomatis infection. Seropositivity for IgM [34.1% (15/44)] as well as IgA [40.9% (18/44)] significantly correlated to PCR positivity, while significant correlation was not seen with IgG positivity. The overall seropositivity (IgM/IgA/IgG) in the study population was 68.5%.

E.Paroli et al[30] studied the seroprevalence of anti - Chlamydia trachomatis IgG antibodies among patients attending STD clinic in Rome, Italy. The population composed of 741 heterosexuals, 470 males and 271 females, and of 147 homosexual-bisexual men. The prevalence rates were

60.0% in heterosexual males, 50.6% in females and 73.5% in homosexuals-bisexuals. A positive association between age and antibody prevalence was found in males. Among heterosexuals there was an increasing trend of seropositivity with number of partners during the previous year.

Malhotra et al[31] from New Delhi studied the prevalence of Chlamydia trachomatis and its association with other sexually transmitted infections in a tertiary care centre. A total of 276 female patients with complaints of genital discharge or ulcer were enrolled in the study. Genital discharge specimens were collected from all patients. The samples were analysed for the presence of antigen and antibody by Direct Fluorescent Antibody test (DFA) and Enzyme Linked Immunosorbent Assay (ELISA), respectively. Chlamydial infection was found in 19.9% of patients.(10.1% by DFA, 10.9% by ELISA). Only three patients (1.1%) had both antigen and antibody positive. The highest rate of infections was found in the age group of 20 – 30 years, the sexually active group.

AIM OF THE STUDY

To determine the seroprevalence of Herpes Simplex virus (HSV1 and HSV2), Hepatitis B and Chlamydia trachomatis among the patients attending the outpatient department of Institute of Venerology, Madras Medical College.

MATERIALS AND METHODS

STUDY DESIGN : Cross-sectional study

SAMPLE : The study population composed of 91patients attending the Outpatient department of Institute of Venerology, Madras Medical College, Chennai from November 2007 to November 2008.

INCLUSION CRITERIA:

Patients attending the OPD irrespective of symptoms.

METHODOLOGY

All patients included in the study were interviewed and data regarding their name, age, address, educational status, occupation and marital status were collected. Their chief complaints, previous illnesses, treatment taken and sexual history were recorded.

A detailed general and systemic examination followed by genital examination was done.

If the patient presents with ulcer, dark field examination for demonstration of Treponema pallidum, smear examination by Gram's

staining for the demonstration of Haemophilus ducreyi and Tzanck smear by Leishman staining for demonstration of giant epithelial cells are done.

In female patients presenting with genital discharge, wet film for Trichomonas vaginalis and KOH preparation for candida were taken.

In all study patients, with informed consent and aseptic precautions, 5 ml blood was drawn and serum was separated by centrifugation and stored in sterile containers under refrigeration at -20^0 C.

All patients' serums were routinely tested for blood VDRL and HIV1 & HIV2 (ELISA).

The serum samples were tested using Enzyme Immunoassay for the determination of IgM and IgG antibodies to Herpes simplex virus type 1 & 2 using Novatec ELISA kits. All reagents and samples were brought to room temperature one hour before use. The procedure of testing was as follows.

- The samples were diluted 1:100 with IgM sample diluents (10 µl sample + 1 ml sample IgM diluents). The controls and

standards were not diluted as per manufacturer's instructions.

- 100 µl of sample and control were then added in the wells. The wells were covered with the foil supplied in the kit.
- The wells were incubated for 1 hour ± 5 min at 37 ± 1° C.
- After incubation, the foil was removed and the contents in the well were aspirated.
- Washing of the wells were done with 300 µl of washing solution. The soak time between each wash cycle is < 5 sec.
- 100 µl of conjugate was added in the wells and incubated for 30 min at room temperature. Again the wells were washed as in the previous step.
- 100 µl of TMB substrate was added and incubated for 15 min in the dark.
- 100 µl of stop solution was added to the wells.

- The adsorbance of each well was read at 450 nm using ELISA reader. The cut off values calculated and results were interpreted.

HBsAg antibody for Hepatitis B was tested using ERBA-LISA kits. The test was done as follows.

- 50 µl of sample diluents were added to each well.
- In each run, there was one blank (100 µl sample diluents + 50 µl conjugate), three negative controls and one positive control.
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- 50 µl of control and test specimens were added to respective wells. 50 µl of conjugate was added to each well.
- The plate was covered with black cover and incubated for 60 minutes at 20 – 37 ^{o}C.
- The plates were washed as per microplate washing procedure.

- 50 µl of colour reagent added, covered with black cover and incubated for 15 minutes in dark at 20 – 30 ^{o}C.

- 100 µl of buffer solution was added to each well and the adsorbance was read at 450 nm.

The serum samples were tested for Chlamydia trachomatis IgG by Enzyme immunoassay using Microwell ELISA kit. The test procedure was as follows.

- 1:21 dilution of test samples were prepared by adding 10 µl of sera to 200 µl of sample diluents and mixed well.

- 100 µl of diluted sera, calibrator and controls were added in appropriate wells and incubated for 20 minutes at room temperature.

- The liquid from all wells were removed and washed three times with 300 µl of 1X wash buffer.

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- 100 µl of enzyme conjugate was added to each well and incubated for 20 minutes at room temperature.

- The enzyme conjugates were removed from all wells and washed as previous step.

- 100 µl of TMB substrate was added and incubated for 10 minutes at room temperature.

- Then 100 µl of stop solution was added.

- The optical density was read at 450 nm using ELISA reader within 15 minutes and cut off values were calculated.

RESULTS

Table 1:- Sex distribution among study patients.

Male	49
Female	42

The study group included 49 males (53.84%) and 42 females (46.15%).

Table 2:- Reason for attending STD outpatient department

Symptomatic	56
Asymptomatic	35

35 patients among 91 in the study group (38.5%) attended STD outpatient department for screening of STD though they were asymptomatic for those diseases.

Table 3:- Age distribution of study patients.

	Hep B		Chlamydia	
	Male	**Female**	**Male**	**Female**
≤ 20	_	_	2	1
21 – 30	2	2	7	6
31 – 40	0	1	4	7
41 – 50	1	_	4	3
51 – 60	_	_	1	_

Age group	**HSV 1**		**HSV 2**	
	Male	**Female**	**Male**	**Female**
≤ 20	1	1	_	2
21 – 30	2	9	9	10
31 – 40	8	8	4	8
41 – 50	3	3	3	3
51 – 60	1	_	1	1

Majority of the patients were belonging to the age group of 21 – 30 years, followed by 31 – 40 years. All the four diseases studied were less prevalent in younger age group i.e. < 20 years and older age group i.e. 51 – 60 years.

Table 4 :- Seroprevalence of Herpes simplex virus 1 & 2 antibodies, HBsAg and Chlamydiae IgG antibodies.

Test	Positive	Percentage
HSV 1 antibodies	46	50.5%
HSV 2 antibodies	41	45.05%
HBsAg	6	6.5%
Chlamydial IgG antibody	35	38.46%

Out of 91 samples tested by each of the six tests, 46 were positive for HSV 1 IgM or IgG or both. Similarly 41 were positive for either one or both of HSV 2 antibodies. 6 were positive for HBsAg ELISA and 35 for Chlamydial IgG ELISA. Hence the prevalence of Herpes simplex virus 1 infection was 50.5%. Similarly the seroprevalence of Herpes simplex virus 2 infection was 45.05%. The seroprevalence of hepatitis B and Chlamydial infection were 6.5% and 38.46% respectively.

Table 5:- Patients with co infections.

Negative for all diseases tested	14
Positive for one disease serology	38
Positive for two diseases serology	28
Positive for three diseases serology	10
Positive for four diseases serology	1

Out of 91 patients tested, 14 were negative for HSV 1 & 2, HBsAg and Chlamydial antibodies tested. Only 38 were positive for one infection. Rest of the patients had co infections.

Table 6:- Patients testing positive for serological test for single disease.

HSV 1 antibodies	17
HSV 2 antibodies	13
Chlamydial antibody	7
HBsAg	1

In patients with single infection, HSV 1 was the predominant. Only one patient was positive only for hepatitis B antibody and negative for the rest.

Table 7:- Patients testing positive for serological tests for two diseases.

Test	Positive	Percentage
HSV 1 + HSV 2	10	10.98%
HSV 1 + Chlamydia	9	9.89%
HSV 2 + Chlamydia	7	7.69%
Hep B + Chlamydia	2	2.19%

10 patients were found to be infected with both Herpes simplex 1 & 2 viruses. Chlamydial infection was found to be coexisting in the rest of 18 patients testing positive for two disease serological tests.

Table 8:- Patients testing positive for serological tests for three diseases.

Test	Positive	Percentage
HSV 1 + HSV 2 + Chlamydia	8	8.79%
HSV 2 + Hep B + Chlamydia	1	1.09%
HSV 1 + HSV 2 + Hep B	1	1.09%

8 (8.79%) patients were found to be infected with both types of Herpes simplex viruses and Chlamydia.

Table 9:- Patients testing positive for the serological tests for all four diseases.

Test	Positive	Percentage
HSV 1 + HSV 2 + Hep B + Chlamydia	1	1.09%

Only one patient was found to be positive for HSV 1 IgM, HSV 2 IgG, HBsAg and Chlamydial IgG. He was also reactive for HIV.

Table 10:- Total no. of samples positive among the tests done.

Tests done		No. of samples positive (Total tested = 91)	Percentage
HSV 1	IgM	15	16.48%
	IgG	39	42.85%
HSV 2	IgM	11	12.08%
	IgG	35	38.46%
HBsAg		6	6.5%
Chlamydia IgG		35	38.46%

Out of the above 6 tests done, the maximum positivity was seen for HSV 1 IgG ELISA (42.85%). 38.46% samples were positive for HSV 2 IgG and Chlamydia IgG antibody ELISA tests. Out of 91 samples tested, only 6 were positive for HBsAg ELISA test.

Table 11a:- Total no. of patients infected with Herpes simplex virus 1 & 2.

Patients with HSV 1 infection only	25
Patients with HSV 2 infection only	21
Patients with both HSV types infection	20
Total no. of patients with HSV infection	67

Out of 91 patients tested, 67 (73.62%) were infected with Herpes simplex virus. Out of these, 37.3% had only HSV 1 infection, 31.3% had only HSV 2 infection and 29.8% had both HSV 1 and 2 infections.

Table 11b:- Antibody type in HSV infected patients.

	IgM + ve	IgG + ve	IgM + IgG +ve
Patients with HSV 1 infection	6	31	9
Patients with HSV 2 infection	6	30	5

Among the patients testing positive for HSV antibodies, majority of them were IgG positive.

Table 12:- HIV positivity among samples tested

	HSV 1		HSV 2		HBsAg	Chlamydia IgG
	IgM	IgG	IgM	IgG		
HIV +ve N = 10	3	4	1	9	2	8

A total of 10 out of 91samples were reactive for HIV by ELISA. All the patients reactive for HIV ELISA were positive for one or more of the six tests done for this study. One patient's sample was positive for HIV and all the other six ELISA tests done for this study.

Table 13:- Co infections in HIV positive patients.

HSV 1 infection	5
HSV 2 infection	9
Hepatitis B	2
Chlamydial infection	8

2 patients had HSV 2 infection alone along with HIV infection. Rest of the patients had mixed infections.

Table 14:- Other associated STIs in seropositive patients.

Associated disease	HSV 1	HSV 2	Hep B	Chlamydia
Syphilis	4	3	_	1
Trichomoniasis vaginitis	_	1	_	1
Bacterial vaginosis	5	6	_	4
Vulvovaginal candidiasis	_	2	_	_
Genital warts	4	1	_	2

Table 15a:- Serological profile of symptomatic patients.

Test	Genital ulcer	Genital discharge
HSV 1	10	10
HSV 2	9	10
Chlamydia	7	8

Out of 46 HSV 1 antibody positive samples, 10 (21.7%) were from patients with genital ulcers. Similarly 21.9% HSV 2 seropositive samples were from patients with ulcers. 8 (22.8%) of Chlamydial antibody seropositive patients had genital discharge.

Table 15b:- Serological profile of asymptomatic patients.

Serological test	Total positive
HSV 1 antibodies	13
HSV 2 antibodies	14
Chlamydial antibody	15

Among the asymptomatic patients, 13 samples were positive for HSV 1 antibodies, 14 for HSV 2 antibodies and 15 for Chlamydial antibody. Out of 35 asymptomatic patients, 5 (14.3%) had HSV 1

infection, 6 (17.1%) had HSV 2 infection, 8 (22.8%) had Chlamydial infection and 9 (25.7%) had co infections.

Table 16:- Patients with genital discharges

Speculum examination	HSV 1 IgM/IgG/ both +ve N = 21	HSV 2 IgM/IgG/both +ve N = 24	Chlamydia IgG +ve N = 17
Cervix healthy	6	4	4
Cervix hypertrophied	_	2	2
Cervical erosions	2	3	3

Speculum examination in females testing positive for HSV 1 & 2 antibodies and Chlamydial antibody revealed a healthy cervix in majority of them. Erosions were seen in 2 (9.5%) females with HSV 1 infection, 3 (12.5%) with HSV 2 infection and 3 (17.6%) with Chlamydial infection.

Table 17:- Urban vs Rural distribution of study patients.

	URBAN		RURAL	
	Male	**Female**	**Male**	**Female**
HSV 1	17	14	8	7
HSV 2	10	15	7	9
Hep B	3	3	_	_
Chlamydia	11	11	7	6

In all the four disease groups, patients from urban areas were predominant than rural areas. Out of patients positive for either one or both of HSV 1 antibodies, 31(67.4%) were from urban areas whereas only 15 (32.6%) were from rural areas. Similarly for HSV 2 antibodies positive patients, 25 (60.9%) were from urban areas and 16 (39.02%) from rural areas. All the 6 patients tested positive for HBsAg were from urban background. The similar trend was seen in Chlamydia antigen positive patients i.e. 22 (62.8%) were from urban areas and 13 (37.1%) were from rural areas.

Table 18:- Marital status of seropositive patients.

	Married		Unmarried		Widow
	Male	Female	Male	Female	
HSV 1	12	19	13	1	1
HSV 2	10	21	7	_	3
Hep B	1	3	2	_	_
Chlamydia	12	14	6	_	3

Married men and women were predominant among seropositive patients. Out of 46 samples positive for either of HSV 1 antibodies, 31 (67.4%) were of married patients. In HSV 2 serology positive patients, 75.60% were married; in HBsAg positive patients, 66.66% were married; in Chlamydial IgG antigen positive patients, 74.3% were married. Among married patients, married women were predominant than married men. Among unmarried patients, all seropositive samples were from male patients except for one female patient positive for HSV 1 IgM antibody. There were no widowers in the study.

The married : single ratio for HSV 1 males 1 : 1.08, females 9.5 : 1; HSV 2 males 1.4 : 1, females 7 : 1; HBsAg males 1 : 2; females 3 : 1; Chlamydia males 2 : 1, females 3.5 : 1.

Table 19:- Occupational status of seropositive patients.

Occupation	HSV 1	HSV 2	Hep B	Chlamydia
Agricultural coolie	1	3	_	1
Driver	3	4	_	4
Housewife	14	11	1	7
Labourer	9	10	1	8
CSW	_	1	2	2
Shopkeeper	5	2	1	2
Watchman	1	1	_	1
Electrician	3	_	_	_
Fruit seller	1	1	_	1
Security	1	1	_	2
Police	1	1	_	_
Student	1	_	_	1
Hotel worker	1	2	1	1
Unemployed	1	_	_	_
Others	4	4	_	4

Both skilled and semiskilled labourers were found to be seropositive in equal proportions. Among the females affected, most were housewives.

Table 20:- Educational status among the test positive patients.

Educational Status	HSV 1 IgM/IgG/both +ve	HSV 2 IgM/IgG/both +ve	HBsAg +ve	Chlamydia IgG +ve
Uneducated	9	8	1	7
1^{st} – 5^{th} std.	10	11	2	10
6^{th} – 12^{th} std.	26	21	3	17
Graduate	1	1	_	1

Most of the patients testing positive for the tests were educated. 56.5% of HSV 1 ELISA positive patients, 51.2% of HSV 2 ELISA positive patients, 50% of HBsAg positive patients, 48.57% of Chlamydial IgG positive patients studied upto secondary school.

DISCUSSION

The seroprevalence of HBsAg, HSV 1 antibodies, HSV 2 antibodies and Chlamydial antibodies among patients who attended the STD clinic were 6.5%, 50.5%, 48.05% and 38.46% respectively.

The seroprevalence of HBsAg in this study was 6.5%. Two studies from a similar south Indian population showed similar seroprevalence. A study by Rajesh et al[17] from Perundurai, Tamilnadu in 105 patients showed a prevalence of 5.71%. A study from Puducherry by Singh et al[18] showed a prevalence of 10% in 270 patients studied. Studies from northern India showed a lesser seroprevalence. The seroprevalence of HBsAg in 497 patients studied by Risbud et al[16] from Pune was 3.6%. Gopi Thawani et al[14] from Kolkata studied 457 patients and the seroprevalence was 3.72%. Jindal et al[15] studied 350 patients in Amristar and the seroprevalence of HBsAg was 3.7%. The difference between each study could be due to number of patients screened and the differences in exposure to the virus among those patients. The difference in seroprevalence was noted in community surveys also. A community cluster survey[32] in Tamilnadu in 1998 screened 1981 healthy blood donors and the community prevalence of HBsAg was found to be 5.7%. A community survey[33] of 20,000 healthy blood donors from different cities in Uttar Pradesh showed a lesser prevalence of 2.25%.

Among 91 patients tested, 67 were found to be infected with Herpes simplex virus. 25 (37.3%) had HSV 1 infection alone and 21 (31.3%) had HSV 2 infection alone. Combined infection was seen in 20 (29.8%) patients. The seroprevalence of HSV 1 and HSV 2 irrespective of single or combined infection was 50.5% and 45.05% respectively.

Among the 91 samples tested for HSV antibodies, HSV 1 IgM was positive in 16.48% samples, HSV 1 IgG was positive in 42.85% samples, HSV 2 IgM was positive in 12.08% samples and HSV 2 IgG was positive in 38.46% samples. Nearly half of the samples tested were positive for IgG of both types of herpes viruses showing that past infection was common than recent infection.

A study by Shivasamy et al[22] from Puducherry showed that 91.5% were seropositive for HSV-1, 85.1% were seropositive for HSV-2, 82.9% were co-infected with HSV-1 and HSV-2, 8.1% were seropositive for HSV-1 alone and 2.2% were seropositive for HSV-2 alone.

A study from Amristar by Aggarwal et al[21] showed a seropositivity of 17.6%. The seroprevalence of HSV 2 in Italy was 24.6% according to

Marco Cusini et al[25]. In Tanzania, it was 42.9% according to Langeland et al[24].

This difference in seroprevalence could be explained by the Smith and Robinson's[34] global review of type-specific HSV prevalence in different geographic areas. According to them, HSV-2 prevalence is highly variable and depends on many factors including country and region of residence, population subgroup, sex and age. HSV-2 prevalence is in general higher among higher risk sexual behavior groups and in women than men. Its seroprevalence is strongly associated with age, increasing from negligible levels in children younger than 12 years to as high as 80% among high-risk population. In a given population and age group, HSV-1 prevalence is almost always greater than HSV-2 prevalence.

In this study HSV 1 seroprevalence is slightly higher than HSV 2 seroprevalence.

The seroprevalence of Chlamydia was 38.46% in this study. The seroprevalence of Chlamydial IgG in Chennai according Joyee et al[29] was 58.7%. A similar study by Malhotra et al[31] showed a prevalence of 10.9%

using antibody detection methods. These differences could be due to the difference in the type of antigens used in the kits and the population studied.

The prevalence of Chlamydia trachomatis infection is found to be differing according to the population studied and diagnostic methods. The prevalence of Chlamydia by antigen testing was found to be 24.5% in Chennai by Joyee et al[35] and 16% by Dowe et al[36] in high risk STD clinic attendees.

Out of 91 patients studied, 14 patients (15.3%) were negative for all the tests. Out of rest positive, 38 patients (41.7%) were positive for single infection. Rest 39 (42.8%) had mixed infections. HSV 1 and HSV 2 were the most common mixed infection which was seen in 10 patients. HSV 1, HSV 2 alone or both infections were seen in 37 out of 39 patients with mixed infections. Herpes simplex virus infection was the most common sexually transmitted infection in this study. A study by Kavina BK et al[37] from Ahmedabad concluded that herpes infection was found to be the most common STD among patients attending STD clinic.

Out of 6 patients positive for HBsAg, one patient was positive for hepatitis B alone, the rest had mixed infections.

The HIV seropositivity was 10.9% in this study. Except for 2 patients, remaining 8 patients had mixed infections. HSV 2 infection was found in 9 out of 10 patients. Chlamydia infection was seen in 8 HIV positive patients.

Wald A et al[38] had suggested that HSV-2 infection could significantly enhance the rates of sexual transmission and acquisition of human immunodeficiency virus in developing countries. Similarly HSV 2 infection was significantly higher in HIV positive patients.

The second most common infection in HIV positive patients was Chlamydial infection. Out of 35 patients with Chlamydial infection, 8 (22.8%) were HIV positive. A similar study in Chennai by Joyee et al[39] had reported 29.5% of the Chlamydia infected patients were HIV positive.

Co infections of STI were seen in 42.8% of patients studied. Among patients with two infections, HSV 1 and HSV 2 co infection was common and was seen in 10.98% of cases. In patients with three

infections, co infections of HSV 1, HSV 2 and Chlamydia was the commonest. Out of the 91 patients studied, only one had all the four STI studied viz. HSV 1, HSV 2, Chlamydia, Hepatitis B.

A study by Thawani G et al[14] in Kolkata also reported that 34.7% of the patients had co infection of STI in their serological survey of STD clinic patients. Infection with one STI increases the risk of acquiring another STI.

HSV 1 and HSV 2 co infection was seen in 20 of 67 seropositive patients. Out of 41 patients who were seropositive for HSV 2, 20 (48.8%) had HSV 1 infection. HSV 1 antibodies were said to be protective against HSV 2 infection. But the higher prevalence of HSV 2 in patients with HSV 1 antibodies was noted in this study. In a study by Fujie Xu[41] from US also had found that approximately 76% of persons who had HSV-2 antibody also had HSV-1 antibody. They concluded that the seroprevalence of HSV-1 and age at infection may influence the epidemiology of clinical genital herpes, even if prior HSV-1 infection does not prevent HSV-2 infection.

In this study the co existence of HSV and Chlamydia was seen in many patients. Deka S et al[40] had said that HSV 2 co infection stimulates the formation of persistent chlamydiae. Their study suggested that HSV attachment and entry can provide the necessary stimulus to alter C. trachomatis development.

Most of patients were in the age group of 20 – 30 years, followed by age group of 30 – 40 years. This younger age predominance was seen in both males and females. Many studies had found that the sexually active age group of 20 – 40 years had more STIs.

Thawani G et al[14] had found that maximum infection was seen in 15-30 years age group, followed by 30-45 years age group and > 45 years age group. Chopra et al[19] studied the seroprevalence of HBsAg and found that 80% of the cases were in the age group of 25-34 years while only 20% were in between the age of 35-44 years.

Aggarwal et al[21] had reported that 45.5% of the HSV seropositive patients were between 21 and 30 years. The similar picture is seen in this study group. HSV 2 seroprevalence was higher in males and females of 21 – 30 age groups.

Among the 35 asymptomatic patients, only 7 (20%) were negative for serological tests of Herpes simplex virus and Chlamydia. 13 of 35 patients had HSV 1 infection and 14 of 35 patients had HSV 2 infection. Aggarwal et al[21] also had found 24% prevalence of HSV 1 and 2 antibodies in asymptomatic females.

The seropositivity of STIs was only slightly higher in married men than unmarried men. But the difference was large in married vs. unmarried women. Ray et al had identified that there was increase in the incidence of STI among married patients than single patients in their retrospective analysis of patients over 15 years. Their married females had 4.5 times higher incidence of STIs than single females. Saikia et al[42]

in their study of STDs in Assam had found that prevalence of STIs among married vs. unmarried men was in the ratio of 1 : 1.15 and in females was 2.3 : 1. Among the married individuals, extramarital sexual relation to the extent of 68% was observed in their study.

Majority of the seropositive patients in this study were educated and had upto secondary level of education. Only 14% of the STD patients were uneducated and 36.4% studied upto secondary school in Bangalore city according to Ramesh K et al[43] in their study of STDs in Bangalore city. In a study by Ray et al[20], 46.5% of patients had secondary education.

Approximately 60% of patients with HSV and Chlamydial infection and 100% of patients with hepatitis B infection were from urban background. This may be due to the study being done in an urban hospital setting or lack of health awareness among urban slum and migrant populations.

No specific occupational predominance was noted in this study. The seroprevalence was more or less equal in unskilled and semiskilled labourers. Many of the affected females were housewives. The similar pattern was seen in a study by Ray et al[20] in Delhi in which 66.2% of male patients were semiskilled labourers and 85.8% of females were housewives.

CONCLUSION

- The seroprevalence of HBsAg is 6.5%, Herpes simplex virus 1 antibodies is 50.5%, Herpes simplex virus type 2 antibodies is 45.05% and Chlamydial antibody is 38.46%.

- Most of the HSV antibodies are of IgG type indicating past infection.

- Infection with one STI increases the risk of acquiring other STIs. 42.8% of the patients studied had co infections.

- HSV 1 and HSV 2 are the most common co infections.

- The seroprevalence of HIV in this study is 10.9%. HSV 2 and Chlamydia are the most common co infections in patients with HIV.

- The prevalence of HSV 1 and 2 antibodies and Chlamydial antibodies are high in asymptomatic patients.

- Most of the seropositive patients in this study are married, educated upto secondary school and urban residents.

PROFORMA

Name: Age: Sex:

STD no:

Address:

Occupation:

Educational status: uneducated / up to 5th / 6 – 12th / graduate

Marital status: married / unmarried / widow / widower

Complaints

- Ulcer
- Discharge
- Others

If ulcer genitalia / H/O previous ulcer genitalia

- Duration of the ulcer
- H/O evolution of the ulcer
- Prodromal symptoms: present / absent
- H/O recurrences: present / absent

- No of recurrences per year

- Any similar complaints in the partner

PAST HISTORY

- Past H/O any venereal disease
- H/O jaundice
- H/O blood transfusion
- H/O surgery

TREATMENT HISTORY

PERSONAL HISTORY

- Smoking / alcohol / tobacco chewing
- H/O intravenous drug abuse

MENSTRUAL HISTORY

LMP Regular / irregular cycles

OBSTETRIC HISTORY

- Total no. of children / Last child birth
- H/O abortion - If yes – 1) spontaneous / induced 2) duration of gestation
- H/O ectopic pregnancy / infertility / sterilization

SEXUAL HISTORY

- H/O recent exposure
- Marital contact
- Premarital contact
- Extramarital contact

SEXUAL ORIENTATION

Heterosexual / homosexual / bisexual

GENERAL EXAMINATION

Pallor / icterus / cyanosis / clubbing / lymphadenopathy

Pulse rate / BP

SYSTEM EXAMIANATION

- CVS
- RS
- ABDOMEN
- CNS

- SKIN AND MUCOSA
- MUSCULOSKELETAL EXAMINATION

EXAMINATION OF GENITALIA

FEMALES

- Inguinal nodes
- External urethral meatus
- Speculum examination:
- Cervix – Healthy / Hypertrophied / Erosion

- Discharge – scanty / moderate / profuse

  Mucoid / mucopurulent / purulent

  Foul smelling

- Ulcer – single / multiple, painful / painless, soft / indurated, bleeds on touch

- Skin and mucosa

- Bones and joints

MALES

- Circumcised / uncircumcised

- Ulcer – single / multiple, painful / painless, soft / indurated, bleeds on touch

- Uretheral discharge / subprepucial discharge

- Inguinal nodes

- Skin and mucosa

- Bone and joints

INVESTIGATIONS

- Blood VDRL
- TPHA
- ELISA for HIV

Genital discharge

- Wet film for TV
- KOH for Candida
- Gram stain
- Uretheral smear (males) – Gonococci
- Vaginal and endocervical smear – clue cells / candida / lactobacilli / Gonococci

Genital ulcers

- Dark field for Treponema pallidum
- Tzanck smear
- Leishman's stain for Donovan bodies

Consent

I am willing for clinical examination and blood sampling for serology of HSV 1 & 2, hepatitis B and Chlamydia antibodies. The procedures have been explained to me.

Date: Signature:

Name:

Blood for

- HBsAg
- HSV1 & HSV2 IgM & IgG antibodies
- Chlamydia IgG antibodies

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