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## **A study to assess the prevalence of Herpes Simplex Type 2 (HSV-2) infections in pregnant women in a tertiary care hospital**

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**Abstract**---Aim: Assess the prevalence of Herpes Simplex Type 2 (HSV-2) infections in pregnant women in a tertiary care hospital. Methods: There were a total of one hundred pregnant women that participated in this research. 5 ml blood samples were obtained using vein puncture and placed in a sterile plain tube. These samples were then allowed to clot for 30 minutes before being centrifuged at 3000 rpm for 5 minutes. Each of the resulting serums was placed in a fresh vial, given a label, and kept at a temperature of -20 degrees Celsius until it was time for the ELISA analysis. In order to determine whether or not the serum included HSV-2 IgG antibodies, an HSV-2 specific IgG ELISA test kit was used. Results: In this research, 100 pregnant women were recruited. HSV-2 infection was found in 40% of people. The largest infected group was pregnant women under the age of 25, with 57.14 percent infected, and the lowest in older age groups. Housewives had the greatest prevalence (71.79 percent), followed by private workers (32%), while government employees had the lowest seroprevalence (8.33 percent). The relationship between viral infection and employment was statistically significant ( $p \leq 0.05$ ). There was no statistically significant relationship between pregnancy stage and HSV-2 infection ( $p > 0.05$ ). Conclusion: HSV-2 prevalence was found to be 40%. As a result, it is critical to test pregnant women for the likelihood of HSV-2 infection so that it may be reduced by adequate treatment.

**Keywords**---Herpes Simplex Type 2 (HSV-2), pregnant women, infections.

## **Introduction**

Infection with herpes simplex virus type 1 (HSV-1) or 2 (HSV-2) during pregnancy may sometimes result in neonatal herpes, which can have potentially life-threatening consequences. Neonatal herpes can occur in infants born to women who have had a long-term HSV infection and also those who acquire the infection during pregnancy. The risk of neonatal herpes is higher among pregnant women who are seronegative for HSV-1 or HSV-2 and who acquire their first (primary) HSV infection during gestation<sup>1</sup>. Factors influencing the maternal to foetal transmission are type of maternal infection (primary Vs recurrent), maternal antibody status, duration of rupture of membranes, integrity of mucocutaneous barrier and mode of delivery (vaginal Vs Cesarean section). Primary HSV infection account for half of the neonatal herpes morbidity and mortality. The complications range from local disease involving skin, eye or mouth, central nervous system involvement to disseminated disease. This higher risk is probably caused by greater viral shedding during initial infections and an inadequate amount of time for mothers to build antibodies that protect their infants from the illness<sup>2</sup>. Throughout the course of medical history in United States, the most common cause of newborn herpes has been HSV-2<sup>3-6</sup>. However, HSV-1, which is often linked with orolabial lesions, is rapidly being recognised as the source of newborn herpes in the United States and around the globe. In certain communities, HSV-1 is responsible for up to 73 percent of cases of neonatal herpes<sup>7-9</sup>. This coincides with a rising percentage of genital HSV-1 infections, which may be the consequence of an increase in the number of teenagers who do not possess HSV-1 antibodies at the time of their first sexual experience<sup>10-12</sup>.

HSV infections in pregnant women are commonly asymptomatic and regular HSV screening in pregnant women is not as a routine practice, the available data about HSV infections in pregnant women are limited. Of all the HSV infection neonatal infection remains the most amenable form for prevention as the infection is acquired at the time of child birth and not during the early gestational period. In this study we are try address the knowledge gap of occurrence of HSV infection in pregnant ladies presenting to our tertiary care hospital.

## **Material and Methods**

The study protocol was approved by the institute ethics committee. This research project was carried out in the department of Microbiology. Study population was the pregnant females attending the hospital for pregnancy related services. A total of one hundred pregnant women that participated in this research. Prior to the collection of the samples, a structured questionnaire was used to gather information on the participants' socio-demographic characteristics. Informed written consent was taken from all the participants before enrolling into the study.

## **Methodology**

Under aseptic precautions, 5 ml blood samples were obtained using vein puncture into a sterile plain tube. These samples were then allowed to clot for 30 minutes before being centrifuged at 3000 rpm for 5 minutes. Each of the resulting

serums was transferred into a fresh vial, given a label, and stored at -20 degrees Celsius till taken up for ELISA analysis. HSV-2 IgG antibodies status was assessed using an HSV-2 specific IgG ELISA test kit.

### **ELISA technique**

Desired number of coated strips were placed into the holder, a 1:40 dilution of each was prepared by adding 5  $\mu$ l of each of the sample, negative control, positive control, and calibrator to 200  $\mu$ l of the sample diluents and mixing it well. The calibrator was used to ensure that the sample was accurate. In each of the relevant wells, one hundred microliters of the diluted sera, calibrator, and control were distributed. In order to create the reagent blank, 100  $\mu$ l of the sample diluents were dispensed into the well labelled 1A. A light tap was given to the holder in order to eliminate air bubbles from the liquid content that was contained inside the wells. The sample was allowed to incubate at room temperature for a period of thirty minutes. After the time for incubation had passed, the liquid content was withdrawn from each of the wells, and 100  $\mu$ l of the washing buffer was used to ensure that the wells were clean. After giving the mixture a quick shake, it was spread out on a pad made of tissue paper. This process is carried out three times in total. After completing the washing stage, 100  $\mu$ l of enzyme conjugate was dispensed into each well, and the test was then allowed to continue incubating at room temperature for another 30 minutes. After the incubation, the enzyme conjugate was thrown away, and the phase of washing up was carried out once again using the washing buffer. After this step, 100  $\mu$ l of the chromogenic substrate tetramethyl benzidine (TMB) was dispensed into each well, and the mixture was left to incubate at room temperature for thirty minutes. Following this, 100  $\mu$ l of 2N HCl was added to each well in order to put an end to the reaction. At 450 nm, an absorbance reading was taken of the contents of the microplate using a device called a microwell reader.

### **Statistical analysis**

A descriptive statistical analysis, carried out using SPSS version 25.0, was performed on the data that were acquired. The Chi-square test was employed as the statistical method to establish association, and the values that were produced were regarded to be statistically significant when the p-value was less than 0.05.

### **Results**

In this research, 100 pregnant women were recruited. HSV-2 infection was found in 40% of people. There was no statistically significant relationship between viral infection seroprevalence and subject age ( $p > 0.05$ ). The largest infected group was pregnant women under the age of 25, with 57.14 percent infected, and the lowest in older age groups. Housewives had the greatest prevalence (71.79 percent), followed by private workers (32%), while government employees had the lowest seroprevalence (8.33 percent). The relationship between viral infection and employment was statistically significant ( $p \leq 0.05$ ). In this research, educational level was not observed to be connected with viral infection seropositivity among pregnant women ( $p > 0.05$ ). It was greatest (62.5 percent) among women with no formal education and lowest among women with higher education (24.5 percent).

There was no statistically significant relationship between pregnancy stage and HSV-2 infection ( $p > 0.05$ ). The prevalence was greatest (47.62 percent) among pregnant women in their third trimester and lowest among women in their first trimester (27.78 percent).

Table 1  
Seroprevalence rate

	Number of women	Positive %
HSV-2	100	40

Table 2  
Seroprevalence of HSV-2 infection among pregnant women

Risk factors	Number of women=100	Number=40	%	p value
Age (Years)				
Below 20	14	8	57.14	
25-35	57	23	40.35	0.88
35-45	22	6	27.27	
Above 45	7	3	42.86	
Occupation				
Government worker	24	2	8.33	
Housewives	39	28	71.79	0.0001
Private worker	25	8	32	
Others	12	2	8.33	
Educational status				
Illiterate	12	9	75	
Up to 8 <sup>th</sup>	29	13	44.83	0.06
Up to 12 <sup>th</sup>	40	11	27.5	
Graduate and above	19	7	36.84	
Parity				
1	36	14	38.89	
2	22	9	40.91	0.62
3-4	26	11	42.31	
Above 4	16	6	37.5	
Pregnancy stages (trimester)				
1 <sup>st</sup>	18	5	27.78	0.07
2 <sup>nd</sup>	40	15	37.5	
3 <sup>rd</sup>	42	20	47.62	
Co morbidity				
Diabetic mellitus	16	7	43.75	
Hypertension	20	9	45	

## Discussion

Genital herpes is a global health problem, it is painful condition causing significant physical and psychological distress. Pregnant ladies with genital herpes can also infect their new born leading to neonatal herpes causing significant morbidity and mortality, which is a preventable situation.<sup>14,15</sup>

The epidemiology of genital herpes differs across nations and between groups of people based on demographic and clinical features of the population. HSV-2 antibody seroprevalence is an accurate approach of identifying the epidemiology of this virus. Serological assays using type-specific glycoprotein gG1 and gG2 are more accurate in distinguishing antibodies directed against HSV-1 and HSV-2.

This study's total seroprevalence of 40% HSV-2 infection is unusually high, particularly when compared to comparable studies conducted in other countries. HSV-2 IgG seropositivity was identified in pregnant women of reproductive age who had spontaneous abortions in Nepal at 4.2 percent<sup>16</sup>, 33 percent in Cuba<sup>17</sup>, and 10 percent in Turkey.<sup>18</sup> Higher percentages than those seen in the current research have been recorded in India and other countries, including 49.1 percent in Zimbabwe.<sup>19</sup> 3.95 percent in Kisumu, Kenya<sup>20</sup>, 63.3 percent in Sudan, and 91.1 percent in South Sudan.<sup>14</sup> There was no statistically significant relationship between viral infection seroprevalence and age of the participants ( $p > 0.05$ ). The largest infected group was under the age of 25 (57.14 percent). and the lowest in older age groups. This research is consistent with the findings of Egbagba and Mordi<sup>21</sup>, who found that age did not correspond with the prevalence of viral infection, and Amar et al.<sup>22</sup>, who found that older age groups had the greatest prevalence and younger age groups had the lowest. One probable explanation for this result is because most of the women were infected with HSV-2 as children, therefore marriage and schooling had little influence on the prevalence later in life.

In terms of occupation, housewives had the greatest prevalence (71.79 percent), followed by private workers (32%), others (8.33 percent), while government employees had the lowest seroprevalence (0%). (8.33 percent). The relationship between viral infection and employment was statistically significant ( $p < 0.05$ ) with government employees having least prevalence, probably due to better access to hygienic practices and health care facilities.

In this research, educational level was not observed to be connected with viral infection seropositivity among pregnant women ( $p > 0.05$ ). It was greatest (62.5 percent) among women with no formal education and lowest among women with higher education (24.5 percent). Education has long been recognised as beneficial in many aspects of life. It aids in making educated decisions and locating valuable information on health status and various methods of preventing sexually transmitted illnesses.<sup>23</sup> Paz-Bailey et al.<sup>24</sup> found the same result in their research. There was no correlation between parity and viral infection among the responders ( $p > 0.05$ ). The infection was greatest in pregnant mothers with 3-4 children (42.31%), and lowest in women with more than 4 children (37.5 percent). This conclusion is validated by other research, and it may both directly and

indirectly prolong the length of a relationship with several sexual partners, both of which are independent risk factors. <sup>25</sup>

There was no statistically significant relationship between pregnancy stage and HSV-2 infection ( $p > 0.05$ ). The prevalence was greatest (47.62 percent) among pregnant women in their third trimester and lowest among women in their first trimester (27.78 percent). This study agrees with the findings of Amar et al.<sup>22</sup> in India but disagrees with the findings of Idress and Elhag in Sudan. <sup>14</sup> The patients' history of blood transfusion was not shown to be linked with HSV-2 infection ( $p > 0.05$ ). The frequency of viral seroprevalence was greater in individuals who had no history of blood transfusion (42%) than in those who had a history of blood transfusion (40 percent).

### **Conclusion**

In the current investigation, HSV-2 prevalence was found to be 40%. It is critical to test pregnant women for the likelihood of HSV-2 infection so that it may be adequate treatment. The research emphasises the need for health education and personal cleanliness, which will aid in lowering the risk of infection among pregnant women in the region.

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