Prevalence Of Asymptomatic Genital Herpes Infection In Women Attending The Antenatal Clinics Of The University Of Port Harcourt Teaching Hospital, Port Harcourt, Nigeria.

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Abstract: There is a large population of individuals who have serologic evidence of *Herpes simples virus* infection without symptoms. The aim of this study is to determine the seroprevalence as well as shedding of asymptomatic genital herpes and factors associated with its acquisition in pregnant women. The study was prospective and cross-sectional. A total of three hundred and seventy five (375) pregnant women who had no history of Herpes simplex virus infection were enrolled in the study and given questionnaires covering socio-demographic, parity, occupation, education, habitat clinical and sexual history. Ten millimeter of blood sample was collected from each participant for determination of type specific IgG antibodies by enzyme-linked –immunosorbent-assay (ELISA) (HerpeSelect). HSV-2 shedding was determined using real time polymerase chain reaction (PCR) of swabs collected from the vulva and cervix. The Data collected were entered into Microsoft excel package 2007and were analyzed using a statistical software package, *Epi-info version 6.0.* manufactured by the Centre for Disease Control (CDC), USA in2001. Chi square (x^2) was used to determine the association between HSV-2 viral shedding and seroprevalence. Potential risk factors for HSV-2 antibodies were identified using univariate logistic regression model. These factors were then used in multivariate logistic regression model. Level of statistical significance was set at _< 0.05. Results showed that HSV-2 antibodies were found in three hundred and fifteen (315) 84% of the 375 pregnant women. HSV-2 antigen was detected in seventy five women (20%) by real time PCR. Seropositve results were significantly associated with low viral shedding (x^2=49.56, p=0.01). Multivariate model showed that seroprevalence of HSV-2 was significantly associated with location, (p=0.05), occupation (p=0.01), parity (p=0.001), and number of sexual partners (p=0.001). Conclusion: The results showed a high seroprevalence of HSV-2 among asymptomatic pregnant women in our environment with about one fifth of them shedding the virus in their vagina. There is, therefore, need for voluntary screening, counseling and treatment of pregnant women for genital herpes as part of routine antenatal care.

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Introduction

The herpes simplex virus (HSV) infection has been known since ancient Greek times. There are two types-HSV-type 1and HSV type 2 (Snigh et al., 2005). Genital herpes may be caused by either herpes type-1 (HSV-1) or herpes type-2 (HSV-2), but globally the large majority of cases are caused by HSV-2 (Paz-bailey et al, 2007). HSV-2 is a sexually transmitted pathogen, causing chronic, wide spread infection which is infectious during both its symptomatic and asymptomatic periods (Agabi et al., 2010). It is the commonest cause of genital ulcers worldwide (Brown et al., 2003). The virus shares a common characteristic with human immunoglobulin deficient virus (HIV) by successfully avoiding clearance by the host immune system by enterinost of the studies done on genital herpes in Africa including Nigeria have looked at seroprevalence, but little is known locally about viral shedding in seropositive and seronegative individuals. These groups of persons are important as they constitute those that will readily infect their partners as well vertical transfer to neonates in case of women (Ozoaki et al., 2006) A study carried out in South eastern Nigeria revealed poor knowledge of genital herpes among pregnant women as only 22.6% of them had ever heard of genital herpes (Onwere et al., 2009). There are currently no studies on the prevalence of genital herpes among pregnant women in Nigeria. Pregnant women are central to genital herpes epidemiology as they are sexually active, have reduced immunity and thus acquire, shed and infect their partners and neonates, especially during the last trimester of pregnancy (Nyiro et al., 2011). There is, therefore, need to acquaint these women of the significance of genital herpes.

In over half of horizontal transmission events and in over 70% of vertical transmission, the source contact is unaware of being infected (Pebody et al.,

2004). The clinician must determine when and how to seek a diagnosis of genital herpes in asymptomatic patients and must be able to recognize unusual as well as the more classical manifestation of genital herpes infections in order to arrive at appropriate counseling and treatment decisions. The laboratory can play a key role in this effect (Snigh et al., 2005). The most specific and unequivocal test for herpes viruses is viral culture but has a low sensitivity and also time consuming. Herpes simplex virus polymerase chain reaction (HSV PCR) which amplifies HSV deoxyribonucleic acid (DNA) in the sample is extremely sensitive and is now the test of choice and the gold standard for HSV antigen detection (Snigh et al., 2005).

HSV causes a lifelong infection with unpredictable reactivation and transmission; and detecting antibodies to HSV will play an important role in identifying carriers of this infection particularly among pregnant women and patients attending sexually transmitted disease clinics (Snigh et al 2005; Loutfy and Alam El-Din 2006). Genital herpes infection in pregnant women is of particular importance because of the risk to the fetus and the newborn (Brown et al., 2003). Its role in enhancing human immunoglobulin deficiency virus (HIV) acquisition and transmission makes it an important public health problem. The most severe form of neonatal disease occurs when infection of the mother occurs during the last stage of pregnancy, at a time when the maternal protective antibodies develop much more slowly for passive transfer to the fetus (Quitib et al., 2010). This last trimester of pregnancy is also the period when urinary tract infection of the mother is at its peak (Mordi et al., 2015). The need, therefore, to screen pregnant women for asymptomatic HSV infection as part of antenatal care cannot be overemphasized. The aim of this study, therefore, is to determine the prevalence of asymptomatic genital herpes type-2 infection among pregnant women and identify factors associated with its acquisition.

Methodology: The study which was prospective and cross-sectional was conducted at the University of Port Harcourt Teaching Hospital, Port Harcourt. A total of three hundred and seventy five (375) asymptomatic pregnant women attending antenatal care who duly signed the informed consent form were recruited into the study. This sample size was used for both serology and real time PCR. Participants were selected from women in their last trimester of pregnancy and attending the antenatal clinic at the University of Port Harcourt Teaching Hospital and who have no symptoms confirmed or ascribed to genital herpes such as vesicles, genital ulcers, irritations or vaginal discharge. They are women aged between eighteen (18) years and fifty (50) years. Exclusion criteria were the pregnant women who were below the of eighteen years and those who have symptoms confirmed or ascribed to genital herpes and are on therapy for herpes or other sexually transmitted infections. The non-consenting pregnant women in their last trimester were also excluded.

Anti-HSV-2 antibodies were screened for in the last trimester of pregnancy to diagnose asymptomatic genital herpes infection. A real time PCR was used to determine the shedding prevalence using vaginal swabs. Ethical clearance was obtained from the ethical review committee, University of Port Harcourt Teaching Hospital. A pre-designed questionnaire was used to collect demographic details and risk factors for genital herpes including age, country of origin, years of schooling, number of pregnancies, number of deliveries, contraceptive method used, age at first intercourse, life time number of sexual partners, history of genital herpes and symptoms suggestive of this condition (vesicle ulcers, fissures, and irritations at the genital area, vaginal discharge and dysuria) and history of other sexually transmitted infections. Pretest counseling was done for participants in this study with emphasis on: Client confidentiality; benefit to the participant; Risk of exposure; Reasons for genital herpes screening; Information about the participant risk behavior and transmission to the neonates; Methods of prevention of herpes and other sexually transmitted infections and the implication of positive and negative results.

Specimen collection and handling.

Blood samples were collected aseptically after obtaining an informed consent from each of the pregnant women. Ten millimeter (10ml) of venous blood was collected into a sterile ethylene diaminetetraacetic acid (EDTA) specimen bottle and transported immediately to the University of Port Harcourt Teaching Hospital laboratory for HSV-2 specific IgG serological determination. Plasma was obtained by centrifuging and test done immediately or transferred to a tightly closed sterile container for storage. Separated plasma was refrigerated at 2-8^{0}c. If the assay was not completed within 48 hours, it was frozen at -20^{0}c. Samples were thawed and mixed prior to use.

Vagina swabs (Dacron) were collected by inserting the swab stick about 2cm into the vagina, rotated and withdrawn and the tip of the swab was broken and dropped into a transport medium consisting of phosphate buffered saline in a universal bottle transported to the molecular diagnostic laboratory for real time polymerase analysis.

Specific Igg Determination Assay

Focus Diagnostic HerpeSelect 2 ELISA IgG kit was used. Briefly, polystyrene microwells were coated with recombinant gG-2 antigen. Diluted plasma

samples and controls were incubated in the wells to allow specific antibody present in the samples to react with the antigen. Non-specific reactants were removed by washing, and peroxidase conjugated antihuman IgG was to determine added and reacts with specific IgG. Enzyme substrate and chromogen were added, and the colour was allowed to develop. After adding the stop reagent, the resultant colour change was quantified by a spectrophotometric reading of optical density (OD). Sample OD readings were compared with reference cut-off OD readings to determine results. (Focus diagostics. HerpeSelect 2 ELISA IgG for the qualitative detection of human IgG class antibodies to HSV-2).

HSV-2 Serology Procedure

In the procedure, all samples , control and calibrator were diluted to 1.101 by adding 10ml of each to 1ml of diluent (protein, surfactant and non-azide and calibrator were used for analysis. After one hour of incubation, the ELISA wells were washed three times by using automatic washing machine (washer 430, organ teknika, Australia) (Halpem et al.,2008). Washing was repeated after adding conjugate (peroxide-conjugated goat anti-human IgG) and incubated for 30minutes. A hundred microliter of substrate reagent (tetramethylbenzidine and organic peroxide in buffer) were added to each well and incubated for 10 minutes. Blue color development at this step indicated a positive test result. An equal amount of stop reagent (1ml sulphuric acid) was added and the intensity of color determined using ELISA reader (reader 530, organic teknika, Australia) at 450nm within 10 minutes of stopping the reaction. The ELISA reader was programmed to calculate index values automatically from the optical values of the calibrators. Previous analysis of longitudinal reliability of focus glycoprotein G-based type-specific enzyme immunoassay showed the possibility of sero-conversion (the initial positive test result becoming negative during follow up). This occurred when the mean positive index value of women who seroconverted was relatively low (less than 2.5). More than 87% of positive test results for HSV-2 in their longitudinal analysis have index value greater than 3.0. (Cherpes et al., 2003).

Real Time Pcr Assay.

Procedure for Nucleic acid Extraction: For isolation of HSV-2 nucleic acid from the original clinical material of each specimen, the Qiamp DNA mini kit was used. Briefly the samples were thawed and left at room temperature. 200ul of sample was mixed with 200ul of cell lyses buffer (Qiagen Inc. Santa Clarita, California). The mixture was vortexed, and 2.5 ul of proteinase was added, and the solution was vortexed again and place in a 70^0c heat block for 10 minutes. The tubes were placed in a 95^0c heating blocks for 15 minutes to inactivate the proteinase. 210ul placed into a Qiagen centrifuge column (catalog no.29163). The column was centrifuged at 6000 x g for one minute, and 500ul of AW wash-buffer (Qiagen Inc.) was added and the sample was then centrifuged at 8,000 rpm for 1 minute at 20,000 x g for two minutes. A total of 100ul of preheated 10nM tris was then added and the tube was placed in a 70^0c dry heat block for 5 minutes and centrifuged 6,000 x g for 1minute to elute the DNA. 10ul of the DNA was used for each PCR.

Procedure for real time Taqman PCR

All PCR amplification reactions were performed in a 50ul volume containing 2x Taqman universal Mastermix (Applied Bio-systems) forward primer (45 p-mol/ul), 2.5 p-mole of reverse primer, 5 mole of Taqman probe and 20ul of isolated DNA. The reactions were carried out in a 96-well plate, which were centrifuged for 1 minute at 1,000g at room temperature in a swing-out rotator (Rotina 48R, Hettich, Tuttlingen, Germany) to remove air bubbles in the reaction vessels.

The reaction and detection were performed with an AB1 (Applied Bio-system Inc.) Prism 7500 sequence detection system.

After incubation for two minutes at 50^0c with Uracil N-glycosylase to inactivate possible PCR contaminants from former reactions. The reaction tubes were incubated for 10 minutes at 95^0c to inactivate the Uracil N-glycosylase and for denaturation step. This was followed by 45 cycles of 15seconds denaturation at 95^0c and 60 seconds annealing and extension at 58^0c.

Result interpretation for Real Time PCR

Real-time measurements were taken, and a Ct value for each sample calculated by determining the point at which the fluorescence exceeded a background limit of 0.04.

Data Handling And Analysis

The data collected were entered into Microsoft excel package 2007 and was analyzed using a statistical software package, *Epi-info version 6.0,* manufactured by the Centre for Disease Control (CDC), USA in 2001. Chi square (x^2) was used to determine the association between HSV-2 viral shedding and seroprevalence.

Potential risk factors for HSV-2 antibodies were identified using univariate logistic regression model. These factors were then used in multivariate regression model. Level of statistical significance was set at >_0.05.

Results

The socio-demographic characteristics involving age; education; place of residence; marital status and occupation are shown in table 1. Age group 25-29 years and 30-34years each accounted for 35.2% of the respondents and both constituted the majority.

Majority of the respondents,243 (64.8%) had tertiary education while only 13 (3.5%) had primary education. More than two thirds of the women reside in semi-urban area urban area, 70(20.1) and 221(83.1%) respectively, and majority were married, 348 (92.8%). Most of the respondents were either privately employed, 103 (27.8%) or engaged in trading 102(27.5%).

Results of serology for HSV-2 antibodies and PCR test are shown in tables 2 and 3 respectively. Three hundred and fifteen of the respondents (84.0% C I: 79. 89-87.56) tested positive for HSV-2 specific antibodies. Seventy five (20% C I: 16. 06-24.4) of the respondents shed HSV-2 as evident by positive real time PCR result.

The association between viral shedding and HSV-2 seroprevalence is shown in table 4. Of the 315 seropositive women, 43 (13.7%) shed the virus while 32 (13.7%) of the seronegative women shed the virus. The association between viral shedding and HSV-2 Seroprevalence was statistically significant (x2=49.56, p=0.01).

Seropositive results were significantly associated with low viral shedding (p=0.01).

The relationship between antibodies to HSV-2 and some of the studied socio-demographic, sexual behavior and clinical factors are shown in tables 5 and 6. Multivariate model shows that seroprevalence HSV-2 was significantly associated with location (p=0.05), occupation (p=0.01), parity (p=0.001), and number of sexual partners (p=0.001).

More than half, 209 (57.6%) of the respondents have not heard of genital herpes. Majority of the women, 20 (56.6%) practiced use of condom while more than two thirds of the respondents admitted to past history of multiple sexual partners, 280 (73.3%) and have had more than one pregnancy.

Table 1: Socio-demographic characteristic of the respondents.

Patients Characteristics	Total Number (%) of patients
20-24	11 (2.9)
25-29	132 (35.2)
30-34	132(35.2)
35-39	65 (17.3)
40-44	35(9.3)
Education	
Primary school	13 (3.5)
Secondary school	119 (31.7)
Tertiary	243(64.8)
Place of residence (location)	
Rural area	57 (16.4)
Semi-urban area	70(20.1)
Urban area	221(63.5)
Marital status	
Single	20(5.3)
Married	348(92.8)
Separated	4(1.1)
Occupation	
Public servant	97(26.1)
Private employment	103(27.7)
Trading/business	102(27.5)
Unemployed	69(18.6)

Table 2: Results of HSV-2 serology

HSV-2ELISA	Frequency Number (%)	95% C. I
Positive	315 (84)	79.891-87.5627
Negative	60 (16)	12.437-20.1099
TOTAL	375 (100)	

Table 3: Results of HSV-2 PCR

HSV 2 PCR	FREQUENCY NUMBER (%)	95 % CI
Positive	75 (20.0)	16.0689-24.4122
Negative	300 (80.0)	75.5878-83.9311
TOTAL	375 (100.0)	

Table 4: HSV-2 virus shedding in relation to seroprevalence of HSV-2 among respondents

ELISA RESULT	Result of Real Time PCR		Total
	NO. POSITIVE (%)	NO. NAGATIVE (%)	
Seronegative	32 (53.3)	28 (46.7)	60
Seropositive	43 (13.7)	272 (86.3)	315
TOTAL	**75 (20.0)**	**300 (80.0)**	**375**

X^2 =49.56; P= 0.01

Table 5: Seroprevalence of HSV-2 antibody in relation to socio demographic characteristics of respondents

Patients characteristics	**HSV-2 serology**		**OR (95% CI) (Univariate Model)**	**Multivariate (overall) p-value**
	+ve. No (%)	-ve. No (%)		
AGE			0.1563	
20-24	9(81.8)	2(18.2)	4.5(0.6-11.4)	
25-29	113(85.6)	19(14.4)	6.0(3.3-10.6)	
30-34	111(84.1)	21(15.9)	5.3(3.0-9.4)	
35-39	49(75.4)	16(24.6)	3.1(1.5-6.3	
40-44	33(94.3)	2(5.7)	16.5(3.4-35.3)	
EDUCATION				0.06
Primary sch.	13(100.0)	0(0.0)	Undefined	
Sec. sch.	105(88.2)	14(11.8)	7.5(3.9-14.6)	
Tertiary	197(81.1)	46(18.9)	4.3(2.9-6.3)	
LOCATION				0.05
Rural area	43(75.4)	14(24.6)	3.1(1.4-6.6)	
Semi urban A.	65(92.8)	5(7.2)	13.0(4.-17.2	
Urban Area	184(83.3)	37(16.7)	4.9(3.3-7.6)	
MARITAL STATUS				0.136
Single	20(100.0)	0(0.0)	Undefined	
Married	288(82.8)	60(17.2)	4.8(3.6-6.7	
Divorced	3(100.0)	O(0.0)	Undefined	
Separated	4(100.0)	0(0.0)	Undefined	
OCCUPATION				0.05
Public servant	90(92.8)	7(7.2)	12.9(5.4-14.9	
Private employment	88(85.4)	15(14.6)	5,9(3.1-11.4)	
Trading/business	83(81.4)	19(18.6)	4.4(2.4-8.1)	
Unemployed	50(72.5)	19(27.5)	2.26(1.4-5.2)	

p-values ≤ 0.05 are considered significant. OR: odds Ratio C1: Confident Interval No: Number.

Table 6: Seroprevalence of HSV-2 serology in relation to sexual and behavioral factors of respondents.

Patients characteristics	HSV-2 serology		OR (95% CI) (Univariate Model)	Multivariate (overall) p-value
	+ve. No (%)	-ve. No (%)		
Age at first coitus (yrs)			**0. 067**	
<20	137(86.2)	22.(13.7-10.6)	6.2 (3.7-10.6)	
21-25	95(83.3)	19(16.7)	5.0 (2.8-9.1)	
26-30	77(81.9)	17(18.1)	4.5 (2.4-8.6)	
>30	4(100.0)	0(0.0)	Undefined	
No of Sexual partners				0.001
Single	63 (72.4)	24 (27.6	2.6 (1.5-4. 7)	
Multiple	244 (87.1)	36 (12.9)	6.8 (4.5-10.2)	
No of Times of pregnancy				0.001
1	66 (71.7)	26(28.3)	2.5(1.4-4.5)	
2	77(85.6)	13(14.4)	5.9(2.9-12.1)	
3	35(77.8)	10 (22.2)	3.5(1.5-8.6)	
4	66(97.1)	2(2.9)	33.0(7.5-68.3)	
5	41(93.2)	3(6.8)	13.7(3.7-23.1)	
>5	16(100.0)	0(0.0)	Undefined	
Condom use				0.783
Yes	174 (84.1)	33 (15.9)	5.3 (3.4-8.2)	
No	136 (85.5)	23 (14.5)		
History of STI				0.9754
Yes	66 (84.6)	12 (15.4)	5.5 (2.6-11.7)	
No	225 (83.6)	44 (16.4)		

p-values ≤ 0.05 are considered significant. OR : odds Ratio C1: Confident Interval No: Number.

Discussion

The very high seroprevalence of 84% of HSV-2 among asymptomatic pregnant women in this study implies that a high proportion of the women had been exposed to HSV-2 at sometimes in the past and had developed some level of immunity (Duran et al., 2004). This finding is consistent with high HSV-2 seroprevalence reported from various rural and urban population in Africa: 93.9% in Cotonou, Benin Republic; 84.1% in Younde, Cameroun; 93.95% in Kisumu, Kenya and 87.7% in Ndala, Zambia (Paz-Bailey et al., 2007; Agabi et al.,2010; Nyiro et al.,2011; Munjoma et al., 2010).

In contrast lower seroprevalence have been reported in other parts of Africa: 49.1% in Zimbabwe and 20.1% among pregnant women in Tanzania (Rathore et al.,2010). Outside Africa, a seroprevalence of 63.1% in pregnant women was reported in Turkey (Duran et al., 2004; Looker et al., 2008), while a lower seroprevalence was reported among pregnant women in China (Zang –Shen et al., 2007).

Viral shedding was detected in 20% of the women as evident by positive real time PCR result from the vaginal swab collected from each of them. This percentage represents the women at risk of transmitting HSV-2 to their newborn during delivery leading to neonatal herpes infection (Corey and Wald 2009). The actual prevalence of HSV-2 shedders in pregnancy would actually be higher than this, considering that this represents a single sampling episode, against the knowledge that vaginal shedding of HSV-2 is intermittent (Jan-Ake et al.,2009).

Comparable to our findings, a shedding prevalence of 17.2% was reported among pregnant women delivering at Chris Harry Baragwanath hospital in Sweto, South Africa. A lower shedding prevalence has been reported: 14% in Gabon; (Ozouaki et al., 2006); 9% in USA; (Cherpes et al., 2005); 7.8% in Italy (Jan-ake et al., 2009).

This study showed a significant association between seropositive results and HSV-2 shedding. Among the 60 seronegative women in the study 32 (53.3%) shed HSV-2 through the vagina compared to 43 (13.7%) prevalence among seropositive women. This suggests that the HSV-2 specific antibody has some protective effect, and that some of the seronegative shedders are recent infections yet to undergo sero conversion. It is also observed that a large number of seropositive women do not shed the virus (Ozouaki et al., 2006).

The discordance between seropositive result and viral shedding observed in this study connotes that serological assay may not be a good marker for viral shedding and indeed, PCR is probably a more sensitive and specific test for viral shedding. (Ozouaki 2006).

The disparity in the prevalence of maternal HSV-2 infection between populations in different countries on one hand and between the developed and the developing countries on the other hand, may reflect the different sexual and behavioral practices (Looker et al., 2008). It is likely that seroprevalence would be higher in countries or cultures where polygamy is practiced and also among populations with poor knowledge, attitude and practice of protective sex such as the use of condom (Ozouaki et al.,2006)

This study observed that a large majority (57.6%) of the women have not heard of HSV-2 infection. This is similar to some literature reports of 22.6% awareness level (Onwere et al.,2009).

The high seroprevalence of HSV-2 infection observed in this study could also be due to early exposure to sex in childhood (Cherpes et al., 2005; Ozouaki et al.,2006). Though this study did not demonstrate a significant relationship between age at first coitus and prevalence of HSV-2, It is likely that most respondents would not disclose current age of first sexual exposure. There is mounting evidence suggesting strong association between age at first intercourse and prevalence of HSV-2 infection (Dickson et al., 2007).

This study showed a significant relationship between number of sexual partners and prevalence of HSV-2 among these pregnant women. This is consistent with reports in the literature (Ozouaki et al 2006; Tassiopoulos et al.,2007), and in keeping with the fact that multiple sexual partners is a high risk factor for sexually transmitted infections (Kalichman et al.,2007). A strong association was demonstrated between parity and prevalence of HSV-2 infection among the respondents. This finding is buttressed by other literature reports and can be a direct effect of increasing duration of sexual activity or indirectly, a reflection of multiple sexual partners, both of which are independent risk factors (Kalichman et al.,2007; DesJarlais et al., 2013). Moreover, majority of the women in the study admitted to history of multiple sexual partners, whereas several other studies have identified early sexual exposure and history of other sexually transmitted infections and unprotected sexual intercourse as risk factors for genital herpes infection (Tassiopoulos et al., 2007).

This study did not establish a significant increase in the risk of HSV-2 infection with early age of first sexual exposure; nor with the history of sexually transmitted infections or non-use of condom. Similar findings have been reported in the literature (Rathmore et al.,2010). The failure to demonstrate the association between age at first sexual intercourse , history of sexually transmitted diseases and non- use of condom in this study may be due to the general limitation of eliciting current sexual habits from the respondents using structured questionnaires (Corey and Wald 2009).

Some aspects of the socio-demographic characteristics of our respondents such as occupation and location were significantly associated with increased risk of HSV-2 infection. These factors may reflect the impact of urbanization on the prevalence of sexually transmitted infections as more than two-third of the respondents reside in semi-urban and urban areas.

The age of the women, marital status, and their education status did not correlate with the prevalence

of HSV-2. This finding contradicts some literature reports (Corey and Wald 2009). A possible explanation for this finding is that most of the, women acquired HSV-2 infection early in childhood, such that in later life, marriage and education acquisition had no apparent effect on the prevalence.

This study had some limitations. First, the design was cross-sectional and therefore all the women selected for the study were sampled or tested once. Since HSV-2 is shed intermittently, it is possible to have missed some of the shedders. More than one episode of sampling would be ideal. Secondly, there were no baseline studies on HSV-2 seroprevalence and shedding among pregnant women in our environment to predicate and relate our findings: thus references used were mainly from Gabon, Tanzania, Zimbabwe and South Africa. Thirdly, the structured questionnaire used to elicit sexual history and behavior may not be explorative enough, as most respondents may not divulge their true sexual history and behavior; thus they were adequately counseled before participating, with emphasis on their confidentiality and benefit of the research.

Conclusion

This study has demonstrated a high HSV-2 seroprevalence of 84% among asymptomatic pregnant women in our environment with about one-fifth of the total women shedding the virus in their vagina. Seropositive results were significantly associated with low viral shedding, while living in the urban area, semi-urban, high parity and history of multiple sexual partners were all independently associated with seropositive results. It is, therefore, strongly recommended that there be voluntary screening of all pregnant women for genital herpes infection as part of antennal care.

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