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Original Article

Sero-prevalence of Hepatitis B Virus Infection and its Risk factors among Pregnant Women Attending Antenatal Clinic at Aminu Kano Teaching Hospital, Kano, Nigeria

Ibrahim Adamu Yakasai, Rabi'u Ayyuba, Idris Suleiman Abubakar, Saidu Adamu Ibrahim

Department of Obstetrics and Gynaecology, Bayero University/Aminu Kano Teaching Hospital, Kano, Nigeria

ABSTRACT

Background: Pregnant women infected with hepatitis B virus (HBV) can transmit the infection to their fetuses and newborns. Neonates who contract the HBV have about 90% risk of developing chronic HBsAg carriage (HBsAg: hepatitis B surface antigen) and chronic liver disease. Neonatal immunization interrupts this vertical and perinatal transmission. **Objectives:** To determine the seroprevalence of HBsAg among pregnant women attending the antenatal clinic at Aminu Kano Teaching Hospital (AKTH) and to identify potential risk factors associated with HBV infection. **Materials and Methods:** A case control study was conducted involving a total of 303 pregnant women attending the antenatal clinic at AKTH and 303 nonpregnant women of childbearing age. Blood sample was collected from each woman and the serum tested for the presence of HBsAg using latex rapid agglutination slide test kit (Cal-Tech Diagnostic Inc., USA) in the laboratory of the hospital. Reactive samples were stored at -20°C and further confirmed for HBsAg using enzyme-linked immunosorbent assay (ELISA) kits (Bio-Rad, France). HBsAg-positive samples were tested for hepatitis B e antigen (HBeAg) using ELISA kits (Orgenics, Israel). A pretested, structured questionnaire was used for the collection of sociodemographic data and possible risk factors. **Results:** The prevalence of HBsAg among pregnant women and nonpregnant women were 7.9 and 7.6%, respectively. There was no statistically significant difference in the prevalence of HBsAg in pregnant and nonpregnant women. The presence of HBeAg was statistically significant among both pregnant and nonpregnant women who tested positive for HBsAg. The risk factors associated with HBV infection were blood transfusion, ear piercing, history of an affected sibling with HBV infection, tattooing, and abortion among pregnant women. **Conclusion:** The prevalence of HBsAg in this study was not statistically different in pregnant and nonpregnant women. There was a high level of HBeAg infection among pregnant women who tested positive for HBsAg. History of an affected sibling with HBV infection, tattoo, and abortion were significant risk factors for HBV infection.

KEY WORDS: Africa, hepatitis B, pregnancy, risk factors

INTRODUCTION

Infections due to viral hepatitis are systemic diseases caused by viruses A–E that mostly involve the liver.[1] It is a DNA virus which belongs to the family *Hepadnaviridae*. The virus was first discovered as 'Australian antigen' and later named hepatitis B surface antigen (HBsAg) in the blood of patients. Hepatitis B e antigen (HBeAg) was discovered later as a marker for patients at a high risk for transmission of the disease.[2]

Infection caused by hepatitis B virus (HBV) is a serious public health problem causing about two billion infections worldwide.[3] Transmission is commonly through blood transfusion, blood products, body fluids (urine, semen, sweat, saliva, and tears), use of contaminated needles, vertical transmission (mother to child through infected birth canal), and sexual contact.[1] Neonates born of chronically infected mothers have a 70–90% risk of the infection progressing to a chronic phase.[3]

Most countries in Africa have a high HBV endemicity, with the exception of Morocco and Tunisia, which have intermediate endemicity.[4] A prevalence rate of 10% of HBV was found among pregnant women in Hong Kong,[3] 12% in Taiwan,[5] and17.3% in Burkina Faso.[6]

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Address for correspondence
Dr. Ibrahim Adamu Yakasai,
Department of Obstetrics and Gynaecology,
Aminu Kano Teaching Hospital, PMB3452, Kano, Nigeria.
E-mail: Ibrahimyakasai57@hotmail.com

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Nigeria is classified among the group of countries endemic for HBV infection. Currently about 18 million Nigerians are infected.[7] A prevalence rate of 4.3 % was reported from Port Harcourt,[8] 5.7% from Ilorin,[9] 11.6% from Maiduguri,[10] and 8.3% from Zaria.[11] A seroprevalence of 23.3% was reported among patients attending all clinics at the Aminu Kano Teaching Hospital (AKTH).[12]

When a pregnant woman is infected with HBV, there is a chance she may infect her fetus; 10–20% of women seropositive for HBsAg transmit the virus to their neonates.[13] In women who are seropositive for both HBsAg and HBeAg, mother-to-child transmission is approximately 90%.[14,15] Infected neonates have an almost 90% risk of chronic liver disease (CLD) and also the chance of spreading the disease to siblings and to the community.[13] Similarly, when pregnant women are infected, they constitute a serious health risk not only to their unborn child but also the society at large.[13]

Risk factors include occupational exposure, ethnicity, and history of sexually transmitted infection, blood transfusion, and exposure to hepatitis.[13] The objectives of this study was to determine the seroprevalence of HBV carriers and the risk factors among pregnant women.

MATERIALS AND METHODS

This was a case control study carried out at AKTH, a tertiary health institution in Kano state, Northwest Nigeria. The study population comprised all pregnant women booked for antenatal care at AKTH between January and April 2011. Nonpregnant women of childbearing age attending the gynecology clinic at AKTH were recruited as the control group. Women below the age of 18 years were part of our exclusion criteria and those who knew their status as HBsAg-positive were excluded; the seroprevalence was not calculated in this group. In the control group, women above the age of 49 years and those discovered pregnant were also excluded.

Sample size estimation

The formula for determining sample size for the comparative study was used. Thus the formula:

$$n = (Z\alpha + Z\beta)^2 (P_1q_1 + P_2q_2)$$

$$(P_2 - P_1)^2$$

n = minimum sample size
$Z\alpha$ = Standard normal deviate set at 95% confidence

level = 1.96
$Z\beta$ = Power of the test to detect difference set at 80% confidence level

P1 = Prevalence of hepatitis B surface antigen among pregnant women (obtained from a previous study)[16]
=2.2%

P_2 = Prevalence of hepatitis B surface antigen among nonpregnant women of childbearing age (obtained from a previous study).[17]
=8.9%

q_1 = complementary probability $(1-p_1) = (1-0.022) = 0.978$
q_2 = complementary probability $(1-p_2) = (1-0.089) = 0.911$
n = 179.18129

So, the minimum sample size was 180 in each group.
To account for incomplete responses, all 303 consenting pregnant women and corresponding controls from the gynecology clinic were recruited for the study.

Sampling and data collection

The recruitment was by a simple random sampling method of consenting pregnant women and nonpregnant women of childbearing age, which make it a case control study.

Questionnaires were structured and pretested. The authors and other resident doctors in the antenatal and gynecology clinic administered the questionnaires to the participants after an orientation.

Sociodemographic factors of age, marital status, ethnicity, educational status, and the gestational age of the pregnancy (for pregnant women only) were recorded on the questionnaires.

Potential risk factors including dental procedures, surgical procedures, sharing sharp objects, HBV vaccination status, use of contraceptives, tattooing, unsafe injections, abortion, blood transfusion, and ear piercing were carefully documented.

A blood sample (4 mL) was collected from all participants from the antecubital vein, using plastic disposable syringes. Each sample for a particular participant was coded for easy identification and transferred to a clean test tube and allowed for natural clotting. After clotting naturally, each sample was separated by centrifugation at 1,500 revolutions per minute (rpm). Part of the sera collected was tested for HBsAg using latex rapid agglutination slide test kits (Cal-Tech Diagnostic Inc., USA) in the laboratory of the hospital where the sample was collected.

Reactive samples were stored at -20°C and further reconfirmed for HBsAg using commercially available ELISA kit (Bio-Rad, France) in the laboratory. HBsAg-positive samples were tested for HBeAg using ELISA kit (Orgenics,

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Israel). Ethical approval was granted by the ethical committee of AKTH, Kano.

Data analysis

Data were entered into a personal computer and analyzed using SPSS version 17 computer software. Comparison of categorical variables was done using the chi-square test, whereas *P* value 0.05 or less was considered significant.

RESULTS

During the study period, a total of 303 consenting pregnant women and 326 consenting nonpregnant women from the antenatal and gynecology clinics of AKTH were enrolled, respectively. Twenty-three nonpregnant women from the gynecology clinic were dropped out of the control group because they did not match the age group.

Table 1 shows the sociodemographic characteristics of the subjects. The mean age [standard deviation (SD)] of the cases was 27.6 (5.8) years and the mean age of the controls was 27.0 (5.8). The study showed no significant difference in the distribution of ages between cases and controls ($P=0.189$,). The age group with the highest frequency among cases 106/303 (35.0%) and controls 133/303 (43.9%) was 18 to 24 years, whereas the age group with the lowest frequency among cases 14/303 (4.6%) and controls 14/303 (4.6%) was between 40 and 44 years. The range was 18–44 years.

Majority in both groups are Hausas, and have some level of secondary education. They are mostly housewives. Others in the occupation group include trade like fish farming, cattle rearing, and poultry. The businesswomen among them usually buy goods and sell.

Figure 1 shows the distribution of pregnant women based on the trimester of pregnancy. Twenty-one (6.9%) (21/303) were in the first trimester, 174/303 (57.4%) were in the second trimester, and 108 (35.6%) were in the third trimester.

Twenty-four (7.9%) out of 303 pregnant women tested positive for HBsAg. Fifteen (62.5%) out of the 24 who tested positive for HBsAg were also positive for HBeAg. In the control group, 23 (7.6%) out of 303 gynecologic patients who participated in the study tested positive for HBsAg. Six (26.1%) out of the 23 who tested positive for HBsAg were also positive for HBeAg.

There was no statistically significant difference in the prevalence of HBsAg in pregnant women and nonpregnant women ($P=0.879$).

There was a statistically significant difference of HBeAg presence among the pregnant women who tested positive for HBsAg ($P<0.001$).

There was also a statistically significant difference of HBeAg presence among nonpregnant women who tested positive for HBsAg ($P<0.001$).

Table 2 depicts the seroprevalence of HBsAg among age

Table 1: Sociodemographic characteristics of case and control groups

	Cases		Controls	
	Frequency (n)	(%)	Frequency (n)	(%)
Age group				
18-24	106	35.0	133	43.9
25-29	86	28.4	74	24.4
30-34	77	25.4	63	20.8
35-39	20	6.6	19	6.3
40-44	14	4.6	14	4.6
45-49	0	0.0	0.0	0.0
Total	303	100.0	303	100.0
Ethnicity				
Hausa	216	71.3	207	68.3
Igbo	24	7.9	28	9.2
Yoruba	24	7.9	5	1.7
Others	39	2.9	63	20.8
Marital status				
Married	297	98.2	238	78.5
Single	Nil	Nil	33	10.9
Divorced/separated	6	2	16	5.3
Widowed	Nil	Nil	16	5.3
Marriage setting				
Monogamy	213	70.3	208	68.6
Polygamy	90	29.7	43	14.2
Unmarried	Nil	Nil	52	17.2
Education				
Primary	66	21.8	16	5.3
Secondary	159	52.4	135	44.6
Tertiary	72	23.8	87	28.7
Qur'anic (only)	6	2.0	65	21.4
Occupation				
Housewives	161	53.1	147	48.5
Businesswomen	27	8.9	79	26.1
Professionals	32	10.6	20	6.6
Artisans	20	6.6	3	1.0
Students	54	17.8	6	1.7
Others	9	3.0	6	2.0

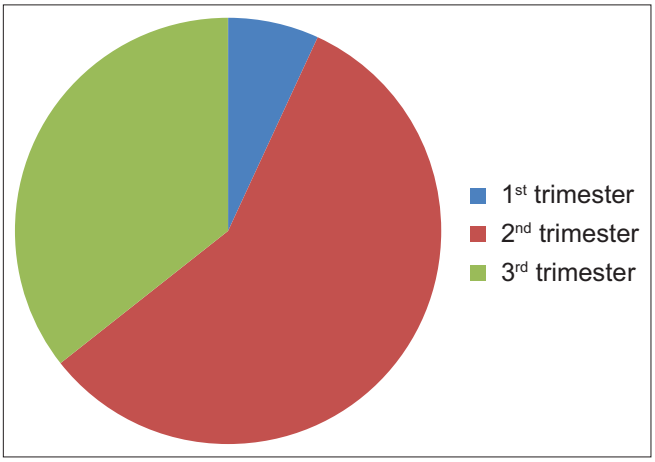

Figure 1: Distribution of pregnant women based on trimester of pregnancy

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groups of both the control and study groups. Except for the age groups of 35–39 and 40–44 years, at least one seropositive case for HBsAg was found in all age groups. The highest age-specific prevalence of 10.5% (9/86) was in the age group of 25–29 years in the study group (cases) whereas in the control group (35.7%) (5/14), it was in the age group of 40–44 years. There was no linear relationship between the prevalence of HBsAg and age in both the study and control groups. There was no statistically significant difference in HBsAg between the age groups among the study group (pregnant women) (P=0.426). However, there was a statistically significant difference in HBsAg between the age groups among the controls (P=0.001).

Hepatitis B surface antigen

Table 3 shows the sociodemographic characteristics of the study and control groups. A high proportion of HBsAg was noted among Igbos (12.5%) (3/24) in the study group. However, there was no statistically significant difference between the ethnic groups (P=0.70). In the control group, a high proportion of HBsAg noted among Igbos (17.9%) (5/28) was statistically significant between the ethnic groups (P=0.017).

An alarming figure of 8.1% (24/297) was also seen among pregnant women who were married, indicating a high prevalence of HBsAg in the group. There was no statistically significant difference of HBsAg between the marital statuses (P=0.468).

In the control group, a high prevalence of HBsAg was noted among the 'separated' women which was statistically significant (P<0.001).

A high proportion of HBsAg seen in the polygamous setting of marriage (16.7%) (15/90) in the study group was statistically significant (P<0.001). In the control group, a high proportion of HBsAg was seen among the unmarried which was also statistically significant (P=0.014).

A high proportion of HBsAg was revealed among those with a secondary level of education (11.4%) (18/159) in the study group. This was however not statistically significant (P=0.141). In the control group, the proportion was highest among those that did not have any form of education (21.4%) (3/14)), which was statistically significant (P=0.040).

HBsAg was more prevalent among professionals/executives (12.5%) (4/32) in the study group but was not statistically significant (P=0.477). In the control group, HBsAg had the highest proportion among the business class, but was not statistically significant (P=0.617).

Eighteen (10.3%, 18/174) pregnant women in their second trimester tested positive for HBsAg, 6/21 (5.6%) pregnant women in their third trimester tested positive for HBsAg; none tested positive for HBsAg in the first trimester of pregnancy. However, occurrence of HBsAg was not statistically significant (P=0.133).

Table 4 depicts the potential risk factors and prevalence of HBsAg among both cases and controls with risk factors. Only tattooing, abortion, and history of an affected sibling with hepatitis B infection among the pregnant women were found to have a significant association with HBsAg (P=0.002, P<0.001, P=0.001, respectively). In the control group, surgical procedure, use of unsterilized sharp instruments, abortion, blood transfusion, ear piercing, and previous infection with HBV were significantly associated with serum hepatitis (P<0.001 P=0.046; P<0.001 P=0.001; P=0.020; P<0.001, respectively).

DISCUSSION

The overall prevalence of HBsAg among pregnant women

Table 2: Prevalence of HBsAg in relation to age of the pregnant women and controls

Age	Cases		Controls	
	No. tested	Positive results (%)	No. tested	Positive results (%)
18-24	106	8 (7.5)	133	11 (8.3)
25-29	86	9 (10.5)	74	3 (4.1)
30-34	77	7 (9.1)	63	2 (3.2)
35-39	20	0 (0)	19	2 (10.5)
40-44	14	0 (0)	14	5 (35.7)
Total	303	24	303	23

HBsAg - Hepatitis B surface antigen

Table 3: Prevalence of HBsAg in relation to sociodemographic characteristics of cases and controls

	Cases		Controls	
	No. tested	Positive results (%)	No. tested	Positive results (%)
Ethnic group				
Hausa	216	21 (9.7)	207	18 (8.7)
Igbo	24	3 (12.5)	28	5 (17.9)
Yoruba	24	0 (0.0)	5	0 (0.0)
Others	39	0 (0.0)	63	0 (0.0)
Marital status				
Single	–	–	33	0 (0.0)
Married	297	24 (8.1)	238	14 (5.9)
Divorced/separated	6	0 (0.0)	16	5 (62.5)
Widowed	–	–	16	4 (25.0)
Educational status				
None	0	0 (0.0)	14	3 (21.4)
Primary	66	3 (4.5)	16	0 (0.0)
Secondary	159	18 (11.3)	135	10 (7.4)
Tertiary	72	3 (4.2)	87	3 (3.4)
Qur'anic	6	0 (0.0)	51	7 (13.7)
Occupation				
Housewife	161	15 (9.3)	147	11 (7.5)
Businesswoman	27	1 (3.7)	79	9 (11.4)
Professional	32	4 (12.5)	20	0 (0.0)
Artisan	20	0 (0.0)	3	0 (0.0)
Student	54	4 (7.4)	43	3 (7.0)
Others	9	0 (0.0)	5	0 (0.0)

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Table 4: Potential risk factors and prevalence of HBsAg amongst the study and control groups

Risk factors	Cases			Controls		
	No.	No. HBsAg$^+$ (%)	*P* value	No.	No. HBsAg$^+$ (%)	*P* value
Dental procedure	59	3 (5.1)	*P*=0.369	50	4 (8.0)	*P*=0.905
Surgical procedure	38	3 (7.9)	*P*=0.995	128	20 (15.6)	*P*<0.001
Sharp instruments	32	0 (0.0)	*P*=0.790	62	1 (1.6)	*P*=0.046
Injectable contraceptive	50	7 (14.0)	*P*=0.082	61	3 (4.9)	*P*=0.378
Loop	42	3 (7.1)	*P*=0.841	47	3 (6.4)	*P*=0.734
Tattooing	93	14 (15.1)	*P*=0.002	42	1 (2.4)	*P*=0.170
Unsafe injection	19	0 (0.0)	*P*=0.187	15	0 (0.0)	*P*=0.255
Abortion	102	18 (17.6)	*P*<0.001	129	19 (14.7)	*P*<0.001
Blood transfusion	40	6 (15.0)	*P*=0.075	100	15 (15.0)	*P*=0.001
Ear piercing	242	19 (7.9)	*P*=0.929	249	23 (9.2)	*P*=0.020
Other risk factors	14	3 (21.4)	*P*=0.550	–	–	–
No risk factors	12	1 (8.3)	*P*=0.957	23	0 (0.0)	*P*=0.153
Miscellaneous	5	1 (20.0)	*P*=0.313	–	–	–
Previous infection	8	0 (0.0)	*P*=0.400	11	6 (54.5)	*P*≤.0.001
Affected sibling	7	3 (42.9)	*P*=0.001	15	3 (20.0)	*P*=0.063
Total	763			932		

was 7.9%, whereas the prevalence among nonpregnant women of child bearing age was 7.6%. These findings were similar to those from Zaria, Northwest Nigeria (8.3%),[11,15] but higher than the findings from Port Harcourt, South Nigeria (4.3%),[18] and Ilorin,[9] North Central Nigeria. However, the prevalence rate in our study was lower than the prevalence found in Maiduguri (11.6%),[9] Northeast Nigeria. The prevalence in both groups also depicts a trend that follows a low prevalence from the southern parts of the country increasing to its highest of 11.6% in the northern parts (Maiduguri). Sociodemographic factors, most importantly the level of health education on prevention, may play a role in the southern parts of the country. Other reasons include higher economic status, higher educational level, early seeking of health-care assistance, and better effective utilization of these health-care facilities.

There was no statistically significant difference of HBsAg in pregnant and nonpregnant women in the study. This could be attributed to similar sociodemographic factors among pregnant women and nonpregnant women of child bearing age in the study and almost equal exposure to the potential risk factors in the study. Pregnancy *per se* is not a potential risk factor for HBV infection.

HBeAg seroprevalence of 62.5% among pregnant women who tested positive for HBsAg and 26.01% among nonpregnant women attending the gynecology clinic at AKTH was alarming. It implies that five out of eight hepatitis-positive pregnant women had HBeAg as did almost one out of four nonpregnant women. As such, there is a high risk of chronic infection and vertical transmission of HBV from mother to child. Although there was no plausible explanation for the large difference in HBeAg rates between pregnant and nonpregnant women, health-care providers attending to these pregnant women should be aware of this high infectivity and provide measures to prevent horizontal transmission. This finding is higher than what was reported by Mbaawuaga and colleagues in Makurdi, North Central Nigeria (3.3%)[19] and 1.39%[10] reported by Harry and colleagues in Maiduguri, Nigeria. The prevalence rate from our findings is also significantly higher than the figure of 0.8% reported from other parts of Africa (Zimbabwe) by Madzime and colleagues.[20] Therefore, the issue of mother-to-child transmission of HBV infection cannot be ignored in AKTH.

The prevalence of HBsAg has no linear relationship in both case and control groups. There was no HBsAg detected in the serum of women in the 35–39 year and 40–44 year groups among pregnant women; all other age groups had HBsAg detected in their serum. A similar finding was also reported by Awole and colleagues[21] in Ethiopia.

The age group of 25–29 years had the highest prevalence of 10.5% in the study group. The high prevalence among this age group was consistently reported in several other studies.[22-24] This is partly because HBV infections are mainly acquired following vertical transmission or through sexual contact, and this group is the most active age group sexually. The positivity rate of HBsAg varies widely among ethnic groups with the highest prevalence recorded among Igbos (12.5%) in the study group and 17.9% in the control group even though this was not statistically significant. A high prevalence of HBsAg was revealed among those who were married in the control group (8.1%). There was no case of HBsAg among the divorced women and the single nonpregnant women. This was similar to the findings of Rabiu and colleagues[25] in Lagos, Nigeria. This could be due to the fact that HBV is sexually transmitted and the duration of sexual activity, number of sexual partners, and history of sexually transmitted infections determine the prevalence of HBV infections.[18]

In the study group, a high prevalence of HBsAg (11.3%) was recorded among those with secondary level of education, similar to the finding of 34 (55.7%) in Lagos. Ndams and colleagues[23] found an inverse association between

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educational status and HBsAg positivity, with less educated women showing the highest prevalence of HBsAg, indicating the positive influence of education and public enlightment/ awareness on the carrier rate of HBV infection.

Pregnant women in the second trimester had the highest prevalence of HBsAg (10.2%), followed by third trimester with a figure of 5.6%. None tested positive for HBsAg in the first trimester. This is similar to the findings of Ndams and colleagues[23] in other parts of Nigeria, where pregnant women in the second trimester had the highest prevalence of HBsAg of 13.4% followed by the third and first trimesters, respectively.

Among the potential risk factors, only tattoo, abortion, and history of an affected sibling with HBV infection among the pregnant women were found to have a significant association with HBsAg. This was similar to the findings in Ethiopia[21,26] where tattooing and abortion were reported as significant risk factors for HBV infection among pregnant women.

The association of HBV infection and abortion could be related to the fact that abortion is directly related to sexual activity and sexually active women have a higher chance of getting the infection especially those in a polygamous setting. A history of an affected sibling with HBV infection implies that the woman was a chronic carrier of the virus and had infected her sibling or the sibling had infected her. Surgical procedure, use of unsterilized sharp instruments, abortion, blood transfusion, ear piercing, and previous infection with HBV were significantly associated with serum hepatitis. In this study, most of the risk factors correlated poorly with the serum hepatitis. This is not an uncommon finding as other studies have indicated that the sensitivity of Center for Disease Control and Prevention (CDC) risk factors for screening pregnant women for HBsAg ranges between 35 and 65%.[27] The findings that only three risk factors among pregnant women were significantly associated with HBV infection clearly indicates the inconsistency of these risk factors and as such, screening pregnant women on the basis of risk factors may be of little help in the detection of HBsAg and prevention of neonatal transmission; hence the need for routine antenatal screening of all pregnant women.

Data on the numbers of excluded women who knew their status as HBsAg positive were not included in the calculation. This may probably imply that the actual seroprevalence of this hospital population may be higher. These shortcomings will be addressed in further studies. Furthermore, some sociodemographic variables which were not included in this study like sexual activity will be given adequate attention and this will include performing a multivariate analysis.

The prevalence of HBsAg among the pregnant women attending antenatal care at AKTH is not significantly different from the nonpregnant women. However, there was a high level of HBeAg among pregnant women who tested positive for HBsAg. Health-care providers should therefore ensure that all efforts are made toward the prevention of vertical and horizontal transmission. These will guarantee optimal feto maternal outcome and healthy health-care workforce, and indeed smoothen the ride toward the developmental goals of the millennium.

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