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SERO-EPIDEMIOLOGICAL SURVEY AND RISK FACTORS FOR HEPATITIS B VIRUS (HBV) INFECTION AMONG PREGNANT WOMEN IN LOGO LGA, BENUE STATE, NIGERIA

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ABSTRACT

Hepatitis B virus (HBV) infection is associated with chronic liver diseases, cirrhosis and hepatocellular carcinoma, liver failure and death. The prevalence of Hepatitis B virus carrier and infectivity status and social characteristics among three hundred and ten pregnant women in Logo LGA, Benue State, Nigeria, was determined through random anonymous testing of volunteers attending antenatal clinics of different hospitals within the community. Thirty of three hundred and ten blood samples tested positive for HBV infection. 11.9% were in the 3rd trimester of their pregnancy and 58.1% were within the age bracket of 21-30 years. Illiterates women constituted 14.4% of those sampled while civil servants were 6.7%. HBV carrier status was determined by the presence of Hepatitis B surface antigen (HBsAg). Repeated reactive samples were confirmed by Enzyme linked immunosorbent assay (ELISA) Kit (Diagnostic Automation, Inc., USA). Maternal HBV infectivity status was determined by testing all HBsAg positive samples for the presence of hepatitis B e antigen (HBeAg). A total of thirty (9.7%) pregnant women identified as carriers of HBV and eleven of the thirty tested positive for HBeAg. Hence, 3.6% (11/310) of the entire study population was found to have high viral replication as well as high risk of transmitting HBV to their neonates. The frequency of HBV carrier did not vary with age, however, it varies significantly with the previous use of contraceptives and the anaemic status of the subjects ($P < 0.05$). This study demonstrates the endemicity of HBV infection in Logo and high infectivity rate, suggest that HBV is likely to be acquired by both vertical and horizontal means of transmission. Testing for HBsAg is recommended for all pregnant women at first prenatal visit so that positive mothers receive prompt intervention.

Key words: Hepatitis B e antigen, Hepatitis B surface antigen, seropositivity, pregnant women, neonates.

ENQUETE SERO - EPIDEMIOLOGIQUE ET LES FACTEURS DE RISQUE POUR L'INFECTION DU VIRUS DE L'HEPATITE B (VHB) CHEZ LES FEMMES ENCEINTEES A LA ZONE DE GOUVERNEMENT LOCAL DE LOGO DANS L'ETAT DE BENUE, NIGERIA.

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RÉSUMÉ

L'infection de virus de l'hépatite B (VHB) est associée aune malade chronique du foie, la cirrhose et carcinome hépatocellulaire, insuffisance hépatique et la mort. La prévalence du porteur de virus de l'hépatite B et le statut de l'infectiosité et de caractéristiques sociales chez trois cent dix femmes enceintes a la zone de gouvernement local de Logo dans l'état de Benue, Nigeria, a été déterminé par des tests anonymes aléatoire des volontaires fréquentant les consultations prénatales des divers hôpitaux à l'intérieur de la communauté. Trente de trois cent dix échantillons de sang ont été positifs de l'infection de VHB. 11,9% étaient au 3^{ème} trimestre de leur grossesse et 58,1% étaient dans la tranche d'âge de 21 - 30 ans. Les femmes analphabètes constituaient 14,4% de celles échantillons alors que 6,7% étaient fonctionnaires. Le statut de porteur de VHB a été déterminé par la présence de l'antigène de surface de l'hépatite B (HBsAg). Echantillons réactifs répétés ont été confirmés par enzymes liée Kit de dosage immun/kit de dosage enzymatique d'immunosorbent lie (ELISA) (Diagnostic Automation, Inc., USA). La maternelle VHB statut d'infectiosé a été déterminée en analysant tous les échantillons HBsAg positifs pour présence de l'antigène de l'hépatite B (HBsAg). Une totales de trente (9,7%) femmes enceintes ont été identifiées comme porteuses de VHB et onze de ces trente ont été testées positives pour HBsAg. Donc, 3,6%(11/310) de l'ensemble de la population d'étude est trouvé d'avoir une réplication virale élevée ainsi qu'une risque élevée de transmettre à leurs nouveau - nés. La fréquence du porteur du VHB ne varie pas avec l'âge, néanmoins, elle varie

de façon significative avec l'utilisation antérieure de contraceptifs et l'état anémique des sujets ($P < 0,05$). Cette étude démontre l'endémicité de l'infection par le VHB à Logo et le taux d'infectiosité élevée, suggèrent que le VHB est probablement d'être contracté par les deux moyens verticaux et horizon taux de transmission. Test pour HBsAg est recommandé pour toutes femmes enceintes à la première visite prénatale pour que les mères séropositives reçoivent une intervention rapide.

Mots - clés : L'antigène de l'hépatite B, l'antigène de surface de l'hépatite B, séropositivité, femmes enceintes, nouveau-nés.

INTRODUCTION

Hepatitis B virus is a blood borne and sexually transmitted pathogen that is spread through percutaneous and mucosal exposure to infected blood and body fluids. The virus was first discovered as "Australia antigen" later named Hepatitis B surface antigen (HBsAg), in patient blood. Hepatitis B e antigen (HBeAg) was identified several years later as a marker for patients at high risk for transmission of the disease (1). The virus has caused several epidemics in parts of Africa and Asia (2) and approximately 350 million persons worldwide are infected with the virus (3, 4, 5, 6), resulting in 2 million deaths annually.

When a pregnant woman is infected with HBV there is a chance she may infect her foetus. It has been reported that 10-20% of women seropositive for HBsAg transmit the virus to their neonates, but in women who are seropositive for both HBsAg and HBeAg; vertical transmission is approximately 90% (7, 8).

Nigeria is classified among the countries highly endemic for viral hepatitis. Currently about 18 million Nigerians are infected (9, 10). Many of these people may not be aware of the infection and hence fail to seek appropriate medical attention therefore progressing to chronic liver disease, cirrhosis and hepatocellular carcinoma, liver failure and death. Similarly when pregnant women are involved they constitute a serious health risk not only to their unborn child but the society at large.

Although, studies have been carried out on HBV in other parts of the country, information is scarce on the prevalence of HBV among pregnant women in the rural parts of Benue state, in particular Logo LGA. The aim of this study therefore is to determine the prevalence of hepatitis B virus carrier and infectivity status of pregnant women attending ante natal clinics in Logo LGA of Benue state, Nigeria.

MATREIALS AND METHODS

AREA OF STUDY

This study/research was conducted in the various health centres within the locality of Logo Local Government of Benue State which is located in an area covering about 1,408 Km² and a population of 169,063 in the 2006 census, in North-Central Nigeria. Its average annual Rainfall is 1200mm and the average annual maximum temperature is 33.3°C. Logo is located on latitude 9°37'N and longitude 6°33'E and it is bordered in the East by Katsina Ala Local Government, in the North by Ukum Local Government, in the West by Taraba State and in the South by Buruku Local Government. Agriculture is the mainstay of its economy with the production of varieties of cash crops throughout the year.

STUDY DESIGN

The study was a hospital based descriptive cross sectional survey conducted between 1st July, 2012 and 30th April, 2013 at the antenatal clinics within Logo LGA, Benue State Nigeria. On every antenatal day, the pregnant women were given health talk on HIV/AIDS and hepatitis infections and were advised on the need to know their status. Only consenting attendees were recruited and included in the study.

SUBJECTS

Three hundred and ten pregnant women attending antenatal clinics in different hospitals in Logo Local Government were randomly selected from the clinic centres. Their specimens were retrieved and reviewed for analysis after informed consent. Information obtained from the case note included demographic characteristics, risk factors, blood group, haematocrit, and results of serological markers for hepatitis B virus.

Pre-structured questionnaires were administered to three hundred and ten consenting pregnant women. Each questionnaire was designed to obtain demographic data such as age, occupation and educational status. Risk factors information were also obtained which include the stages of pregnancy, history of blood transfusion, and history of STIs, whether or not respondents share sharp objects like razors and toothbrushes.

ETHICAL CONSIDERATIONS

Ethical approval was gotten at the Benue state Ministry of Health and Human Services, Makurdi through the Medical Director of the General Hospital Ugba and the Logo Local Government Health Department. Informed consent was obtained from the from the antenatal clinic attendees with assurance that all information obtained would be treated as confidential and would be used for the purpose of this study only.

SAMPLE SIZE AND ITS DETERMINATION

Using the Kish (11) formula: $n = (z^2pq/d^2)$ for determining adequate sample size and further correcting for population less than 10,000 using $n_f = n/1+(n/N)$ (12), 310 respondents were enrolled for this survey.

COLLECTION OF BLOOD SAMPLES / SERUM PREPARATION

Hepatitis B surface antigen (HBsAg) detection was done using the in vitro diagnostic kit manufactured by Cal - Tech Diagnostic, Inc. USA. The test kit (dipsticks) is a rapid immunochromatographic assay designed for qualitative determination of HBsAg in human serum or plasma. Assays were carried out at room temperature. The sera samples were removed from the freezer and left at room temperature to thaw. The test strips were removed from their foil pouches and immersed into serum samples with arrows pointing towards the samples. The strips were taken out after about 10secs and placed on a clean, dry, non-absorbent surface. This is to allow time for the reaction to take place. It was observed that the specimen was absorbed into the test strips and moved by capillary action upward towards the control line. Results were read after 10mins post immersion. Positive samples generated a colour band in the test region of the strips and another in the control region while negative samples had a colour band in the control regions only. It utilizes a combination of monoclonal and polyclonal antigen body to selectively detect elevated level of HBsAg in serum/plasma. The test was carried out and interpreted according to the manufacturer's instructions and in the laboratories of the hospitals where the samples were collected.

Reactive samples were stored in a freezer and further confirmed for HBsAg in the Innovative

Biotechnology Laboratory Keffi where HBsAg positive samples were tested for HBeAg, associated with infectivity and active virus replication; using commercially available enzyme linked immunosorbent assay kit ELISA (Diagnostic Automation Inc. USA)

DATA ANALYSIS

Data from the questionnaires were analyzed using SPSS version 15.0. Chi-square was used to compare significant differences between HBV prevalence and risk factors. Significance was determined at $P < 0.05$ at 95% Confidence interval.

RESULTS

The results of the Seroprevalence study are presented in table 1, showed that of the three hundred and ten (310) pregnant women tested, 30 (9.7%) pregnant women were seropositive for Hepatitis B surface antigen (HBsAg). Eleven (11) out of the thirty (30) pregnant women were identified as HBeAg positive. Hence, 3.6 % (11/310) of the entire study population was positive for HBeAg.

With respect to age, the results showed that there is an increase in HBsAg titres with increase in age up to 30 years followed by a decline. Statistically, however, there was no significant association ($\chi^2 = 1.960$; $p = 0.5808$) between age and seroprevalence of HBV infection.

The level of educational attainment and occupation and seroprevalence of the pregnant women are presented in table 2. It showed that there is an inverse relationship between educational attainment of the women and seroprevalence of HBV infection. Details show that women with high prevalence of the infection are illiterates (14.4%) while those with some levels of education had lower prevalence even though there was no significant association ($\chi^2 = 4.213$ $p = 0.239$). Similarly, the results revealed that house wives had higher prevalence (13%) than the other women considered in this study. Despite this observation, no significant association ($\chi^2 = 5.86$ $p = 0.119$) between HBV infection and the occupation of the women. There was no significant association between infection and pregnancy stages of the women ($\chi^2 = 2.239$ $p = 0.326$), (Table 3).

TABLE 1: AGE SEROPREVALENCE OF HEPATITIS B SURFACE ANTIGEN AND HEPATITIS B E ANTIGEN AMONG THE PREGNANT WOMEN.

Age group	No. examined	HBsAg seropositivity (%)	HBeAg seropositivity (%)
11-15	10	1 (10)	1 (20)
16-20	90	7 (7.8)	4 (4.4)
21-25	100	11 (11)	4 (4)
26-30	80	9 (11.3)	1 (1.3)
31-35	25	2 (8)	1 (4)
36-40	3	0 (0)	0 (0)
41-45	2	0 (0)	0 (0)
Total	310	30 (9.7%)	11(3.5%)

$X^2=1.960$, $p = 0.5808$

$X^2 = 2.222$, $p = 0.5276$.

Education; $x^2 = 4.213$ $p = 0.239$; Occupation; $x^2 = 5.862$ $p = 0.119$

TABLE 2: SOCIAL CHARACTERISTICS AND HEPATITIS B VIRUS SEROPOSITIVITY AMONG THE PREGNANT WOMEN.

Social characteristics	No. examined (n = 310)	No. positive (n=30) (%)
Education		
Illiterate	104	15 (14.4)
Primary	99	5 (5.1)
Secondary	65	6 (9.2)
Tertiary	42	4 (9.5)
Occupation		
Civil servant	15	1 (6.7)
House wife	200	26 (13.0)
Students	17	1 (5.9)
Business	78	2 (2.6)

TABLE 3: HEPATITIS B VIRUS SEROPOSITIVITY AND STAGE OF PREGNANCY AMONG THE PREGNANT WOMEN.

Trimester	No examined	No positive (%)
First trimester (1-3 months)	39	3 (7.7)
Second trimester (4-6 months)	86	5 (5.8)
Third trimester (7-9 months)	185	22 (11.9)
Total	310	30 (9.7)

$X^2 = 2.239$

$p = 0.326$

The frequency of HBV carriers did not vary significantly with blood transfusion, sharing needles, Alcoholic consumption, Herbal medicine use and history of STI(s). However, there was a significant frequency variation in the distribution of HBsAg and contraceptive use subjects ($x^2 = 7.212$ $p = 0.007$) and anaemic and non- anaemic subjects ($x^2 = 7.143$ $df = 1$ $p = 0.008$) (Table 4). HBV carriers also decrease as anaemia decreases significantly ($x^2 = 15.048$ $df = 6$ $p = 0.0199$), (Table 5). Among all the contraceptive devices used previously by the

subjects, only users of oral pills were positive for HBeAg. (Table 6).

DISCUSSION

This study showed that the prevalence of hepatitis B virus (HBV) infection in apparently healthy pregnant women attending antenatal clinics in the major hospitals within Logo LGA, Benue State was 9.7%, falls within figures reported for other African countries. This corroborates the World Health Organization (13) report for Nigeria as highly

endemic area with prevalence greater than 8%. The HBsAg seropositivity of 9.7% among pregnant women in Logo shows that Logo like other areas in Nigeria is endemic for HBV infection.

In related studies in different parts of Nigeria, higher prevalence rates of 13.8% were reported among pregnant women in Lagos (14) 11.6% in Maiduguri (15), 12.6% in North central Nigeria (10) 11% in (16) and 12.3% in Minna (17).

Other findings were, 8.3% in Zaria (18), 2.19% in Benin City (19), 4.3% in Port Harcourt in 2005 (20)

and 2.89% in 2006 (21) and 5.7% in Ilorin (22). The result of this study is higher than the 6.3% reported in pregnant women in Tanzania (23) and 3.7% in Ethiopia (24), showing that variations exist within the same continent of Africa.

Similar studies in other parts of the world were 10% in India (25), 12% in Taiwan (26) and 17.3% in Burkina Faso (27) agreeing with Juszozyk (28) that the global prevalence of chronic HBV infection varies, highest in Africa, Asia and the western Pacific (>8%) to lowest in western Europe, North America and Australia.

TABLE 4: DISTRIBUTION OF HBSAG POSITIVE CASES ACCORDING TO RISK FACTORS AMONG PREGNANT WOMEN.

Risk Factors		No. Examined	Seropositivity (%)	χ^2 , df=1 p value
Blood transfusion	Yes	15	3(20)	1.453 0.228
	No	295	27 (9.2)	
Sharing needles	Yes	270	21(7.8)	0.015 0.0903
	No	40	9 (22.5)	
Contraceptive use	Yes	109	18(16.5)	7.212 0.007
	No	201	12 (6)	
Alcoholic consumption	Yes	105	11(10.5)	0.095 0.758
	No	205	19 (9.3)	
Herbal medicine use	Yes	200	22(11)	0.938 0.333
	No	110	8(7.3)	
Anaemic status	Anaemic	45	10(22.2)	7.143 0.008
	Not Anaemic	265	20(7.5)	
History of STI(s)	Yes	50	5 (10)	0.006 0.939
	No	260	25 (9.6)	

TABLE 5: AGE DISTRIBUTION OF HBSAG AND HBEAG AMONG ANAEMIC PREGNANT WOMEN

Age group	No. examined	Anaemic (%)	HBsAg positivity (%)	HBeAg positivity (%)
11-15	10	6 (60)	1 (10)	0 (0)
16-20	90	14 (15.6)	4 (4.4)	0 (0)
21-25	100	12 (12)	3 (3)	3 (3)
26-30	80	10 (12.5)	2 (2.5)	2 (2.0)
31-35	25	2 (8)	0 (0)	0 (0)
36-40	3	1 (33.3)	0 (0)	0 (0)
41-45	2	0 (0)	0 (0)	0 (0)
Total	310	45(5%)	10(3.2%)	5(1.6%)

$\chi^2 = 15.048$ $p = 0.0199$ $\chi^2 = 2.801$ $p = 0.8334$ $\chi^2 = 15.048$ $p = 0.0199$ $\chi^2 = 2.801$ $p = 0.833$

TABLE 6: DISTRIBUTION OF HBSAG AND HBEAG AMONG PREGNANT WOMEN THAT USED CONTRACEPTIVES PREVIOUSLY.

Contraceptives	No examined	HBsAg positivity	HBeAg positivity
Oral pills	75	14 (18.7)	5(6.7)
Inject able	26	2 (7.7)	0 (0)
Loop	1	0 (0)	0 (0)
Pills/inject able	7	2 (28.6)	0 (0)
Total	109	18 (16.5)	5 (4.6)

$\chi^2 = 4.128$ $p = 0.6594$

HBeAg seroprevalence of 3.6% in the entire study population implies that one out of every three HBV carriers (11/310) is at high risk of transmitting HBV to her neonate as well as higher chances of chronicity. This is alarming but tallies with the result of Olubuyide *et al* (29), who found 3.4% among HBsAg positive doctors and dentist at university teaching hospital Ibadan. However, it is very high when compared with 0.8% found by Madzine *et al*, (30), in Zimbabwe as well as 6.64% and 1.39% obtained by

Harry *et al* (15) among blood donors and pregnant women in Maiduguri respectively. Hence the issue of vertical transmission in sub-Saharan Africa cannot be ignored.

High prevalence of HBV carriers among teenagers (11-25) and 26-30 age groups and the corresponding high HBeAg prevalence among the same age group further show the severity of the infection in the community.

Analyses showed that out of the 310 respondents (33.5%) were illiterates out of which 50% tested positive for HBsAg. There is an inverse association between educational status and HBsAg positivity with less educated women showing the highest positivity, indicating the positive influence of education and public enlightenment/ awareness on the carrier rate of HBV infection. Although HBV infection is considered one of the most important occupational infectious hazards in developed countries (31), result from this study did not reveal statistical significance between HBV seropositivity and the different occupations of the pregnant women studied.

Most of the studied women (185, 59.7%) were in the 3rd trimester of gestation. This group also had the highest HBsAg seropositivity 11.9% (22/185),

followed by those in the 2nd trimester of gestation, results revealed no significant association between infection and pregnancy stages of the women.

The distribution of HBsAg and HBeAg among teenagers and 26-30 years age groups as shown in table 4 and 5 could be due to anaemia which significantly decreases with increasing age. It could also be attributed to the effect of oral pills (6) towards higher expression of HBeAg (Table 1) among the teenagers and 26-30 age groups. Oral pills are steroid hormone prepared in tablet form and have slower but longer action in the body stimulating the immune system for a longer period of time. They have been reported to immunosuppress (blind) lymphocytes by reducing the reactivity of T-lymphocytes and reduce immunoglobulin secretion (32; 33). Hence anaemia, effect of oral pills and other unanalyzed factors such as HIV and malaria may have contributed immensely to the endemicity of HBV infection among pregnant women in the study community, Logo LGA.

CONCLUSION

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Since the virus can be transmitted from infected mother to the offspring especially at birth, the presence of HBV infection in women in general and pregnant women in particular calls for concern. Free screening of all pregnant women should be incorporated in the antenatal and post-natal infection programmes in hospitals to prevent potential infection of the infants by their infected mothers. The use of other HBV infection serological markers such as anti HBs, anti HBc, and HBe as well as HIV co-infection and cases of HBsAg positive women and the effects of the positivity on their babies is advocated for further studies.

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