Archives of Immunology and Allergy

ISSN: 2639-1848

Volume 1, Issue 2, 2018, PP: 67-74

Prevalence of Hepatitis D Virus Antigens Among Sero-Positive Hepatitis B Surface Antigen (HBsAg) Patients Attending Aminu Kano Teaching Hospital (AKTH), Kano

Waiya S.A[1], Taura, D.W[2], Shehu A. A[3], Yahaya S.M[4], Ali M*[5] and Garba M[2]

[1]School of Hygiene, Kano.

[2]Department of Microbiology, Bayero University Kano.

[3]Department of Microbiology, Kano University of Science and Technology Wudil.

[4]Department of Biochemistry, Kano University of Science and Technology Wudil.

[5]Department of Microbiology, Federal University, Gusau.

alimuhd4real@gmail.com

***Corresponding Author:** Muhammad Ali, Department of Microbiology, Federal University, Gusau, Nigeria.*

Abstract

Hepatitis D virus (HDV) is a defective RNA virus presenting similarities to some plant viroids, which requires hepatitis B virus (HBV) as a helper virus for its propagation.The study was aimed to determine the prevalence of Hepatitis D virus antigens among sero-positive hepatitis B surface antigen (HBsAg) patients attending Aminu Kano Teaching Hospital (AKTH), Kano.A total of 115 patients with various forms of liver disease were screened for HBsAg. Results had shown that 92 (80%) tested positive for HBsAg. These were made up of 10 patients with acute hepatitis (10.9%), 15 patients with asymptomatic infection (16.3%), 5 patients with chronic hepatitis (5.43%), 20 patients with liver cirrhosis (21.73%) and 42 patients with primary liver cell carcinoma (45.65%). There were 57 males (61.96%) and 35 females (38.04%).The result of distribution of hepatitis D among hepatitis B subjects showed that 5 (5.43%) out of 92 subjects have hepatitis B and D co-infections. Age category 21-30 and 31-40 years have the highest co-infection with 2 subjects in each case while 41-50 years has 1 subject.The prevalence of Hepatitis D virus with respect to sex among Hepatitis B subjects higher among female with 3 subjects accounted for 60% than male with 2 subjects (40%).Statistical analysis of the result showed significant different in the prevalence of hepatitis B and D at probability level of p<5% (0.05%).

Keywords: *Antigen, Hepatitis B, Hepatitis D, Prevalence, Virus.*

Introduction

Hepatitis D virus (HDV) is a defective RNA virus presenting similarities to some plant viroids, which requires hepatitis B virus (HBV) as a helper virus for its propagation. Among 240 million chronic HBV carriers reported worldwide [1], approx. 15 to 20 million individuals are also infected with HDV [2,3]. In Africa, of the estimated 65 million chronic HBV carriers, about one fourth of HBsAg-positive individuals show dual-infection with HDV [3]. Chronic HBV/HDV co-infection can lead more often to severe liver diseases, like fulminant hepatitis, if compared to HBV mono-infection [4], whereas HBV/HDV super-infection is associated with chronic infection among 90 % of the virus carriers [5].

Hepatitis D virus (HDV) is a spherical hybrid particle of ~36 nm in diameter, composed of an outer coat containing hepatitis B surface antigens (HBsAg) and host lipids. The inner nucleocapsid consists of small and large hepatitis D delta antigens (sHDAg and LHDAg) and a single-stranded, circular RNA molecule of ~1.7 kb [6]. The unique open reading frame of the HDV genome encodes the delta antigens. sHDAg is required for HDV genome synthesis while LHDAg inhibits HDV-RNA synthesis and is essential for HDV particle formation [7]. Currently, eight HDV genotypes

(HDV1-8) have been described with variable geographical distribution [8,9].

HDV1 is the most common genotype that distributes globally [8,10]. HDV2 is prevalent in Asia while HDV3 is detectable in South America. In sub-Saharan countries, the prevalence of HDV1, HDV5, HDV6, HDV7 and HDV8 has been reported previously [3,11]. Clinically, HDV1 infection is associated with both severe and mild liver disease, whereas HDV2 and HDV3 are associated with mild clinical course and outbreaks of severe fulminant hepatitis, respectively [12].

The HDV prevalence has been reported in various parts of the world. In Africa, the anti-HDV antibody prevalence in HBsAg carriers was reported only in Cameroon (17.6 %) and Gabon (15.6 % to 70.6 %)[11,13,14]. Recently, HDV prevalence in sub-Saharan Africa was estimated from 1.3 % to 50 % [3]. Although HBV is endemic in Nigeria, data on HDV sero- prevalence are limited. A previous study showed that HDV antigen was detectable in 6.5 % of patients with chronic hepatitis B in Southwest Nigeria [15]. In addition, another study reported an anti- HDV prevalence of 12.5 % in 96 HBsAg positive patients [16]. Moreover, a recent study showed that HDV1 prevails with 53.3 % in Southwestern Nigeria followed by the HDV5 (33.3 %) and HDV6 (13.3 %), which were more restricted to the northern part of Nigeria [3]. The study was aimed to determine the prevalence of Hepatitis D virus antigens among sero-positive hepatitis B surface antigen (HBsAg) patients attending Aminu Kano Teaching Hospital (AKTH), Kano

Materials And Methods

Ethical Approval

The ethical clearance for the study was approved by the research ethics committee of Aminu Kano Teaching Hospital (AKTH) Kano and informed consent was also obtained from all the participants (subjects).

Study Area

The study was conducted at **Aminu Kano Teaching Hospital (AKTH)** Kano. Kano state is located in the North-west Nigeria with coordinates 11^0 30 N 8^0 30 E. It shares borders with Kaduna state to the south-west, Bauchi state to the South-East, Jigawa state to the East, Katsina state to the West and Niger republic to the North. It has a total area of 20,131km^2 (7,777sqm) and population of 11,058,300 [17].

Research Design

This was a prospective, cross-sectional study of involving patients with clinical features of liver disease seen at the Department of Medicine, Aminu Kano Teaching Hospital (AKTH) between June, 2016 and December, 2016. The population study was a heterogeneous population consisting of different age group, ethnicities and educational status. Bio-data and other information of the patients were collected using informed consent forms with the assurance that all information obtained has to be treated with confidentiality.

Sample Size Determination

Sample size for the study was determined from a standard formula for the calculation of minimum sample size [18]. Sample size was given by the formula

$N = (Z_1\text{-}a)^2 (p) (1\text{-}p) / d^2$

N = minimum sample size

$Z_{1-}a$ = value of standard normal deviate which at 95% confidence interval has found to be 1.96.

P = the best estimate of prevalence obtained from literature review (8%) and

d = difference between the true population rate and sample that can be tolerated, this is the absolute precision (in percentage) on either side of the population.

$N = (1.96)^2 (0.08) (1\text{-}0.08)/ (0.05)^2 = 113.35$ as the minimum number of samples for the study.

Therefore, a total of 113 with additional of 2 subjects were added to the research for attrition, making a total of approximately 115 samples.

Samples Collection

A total of 115 blood samples were collected from suspected Hepatitis B patients that attended Aminu Kano Teaching Hospital (AKTH) using blood collection vacutainer tubes and needles in vein puncture. This blood was then transferred into a plain container containing no anticoagulants.

Detection of Hepatitis B Virus (Hbv)

Gold Rapid Screen test was used in this study for the detection of Hepatitis B Surface Antigen (HBsAg) in the blood serum as described by Djebbi et al.

[19]. The blood collected from suspected patients with Hepatitis B was left to clot and the serum was separated by centrifugation. The serum was then carefully withdrawn for testing. Using Pasteur pipette, 70-100µl (2-3 drops) of serum was added into the sample pad and observed for 15 minutes.

Liver Enzymes and Liver Function Test

The hepatitis B disease was evaluated using liver enzyme tests by detecting the level of Alanine Amino-transferase (ALT), Aspartate Amino-transferase (AST), Alkaline Phosphatase (ALP), and liver function tests (LFTs) that include Direct Bilirubin (D_B) and Total Bilirubin (T_B) tests as described by Changotra et al., [19].

Detection of Hepatitis D Virus (HDV)

Hepatitis D Virus was detected using Ag ELISA test.The procedure was conducted according manufacturer's instructions. The reagents were kept at room temperature (25 ^{0}C). The Formed crystals were resolubilized by warming at 37 ^{0}C until crystals dissolved. Wash Buffer 1 to 20 was also diluted with distilled water using clean vessels. The strips needed were set in strip-holder and sufficient number of wells (required for the test) were numbered including three Negative control (B1, C1, D1), two Positive control (E1, F1) and one Blank (A1) which neither samples nor HRP-Conjugate were added into the Blank well. 50µl of Positive control, Negative control, and specimen were added into their respective wells. 50µl of Extraction Solution supplied with the kit was added using separate disposal pipette for each specimen to each well in order to avoid cross contamination and then slightly mixed. The extraction Solution was never added to the Blank Well. The plate was covered with plate cover and incubated for 30 minutes at 37^{0}C. After incubation, the plate cover was removed and discarded. Each well was washed 5 times with diluted Wash buffer. The micro-wells were each allowed to soak for 30-60 seconds. After the final washing cycle, the strips plate was turned onto blotting paper or clean towel to tap out any remainders. Then, 100µl of HRP-Conjugate Reagent was added into each well and mixed gently. The HRP-Conjugate was never added to the Blank Well. The plate was covered with the plate cover and was incubated for 30 minutes at 37^{o}C.After incubation, The plate was washed 5 times with wash buffer for the second time. About 50µl of Chromogen A and 50µl Chromogen B solution were dispensed into each well including the Blank and mixed by tapping the plate gently. The plate was incubated at 37^{0}C for 10 minutes avoiding light. The enzymatic reaction between the chromogen solutions and the HRP-Conjugate produces blue color in positive control and HDV-Ag Positive sample wells. Using a multichannel pipette, 50µl of Stop Solution was added into each well and mixed gently. Intensive yellow color develops in Positive control and HDV-Ag Positive sample wells. The plate reader and the Blank well were calibrated and the absorbance was read at 450nm. The Cut-off value was calculated and the results were evaluated. The absorbance was read 5 minutes after stopping the reaction.

Statistic Analysis

The T-test was performed for quantitative variables to check for relationship of HBV and HDV infection and the significant level for HBV and HDV was set at probability level of $P < 0.05$.

Results

Demographic Distribution of the Subjects and Incidence of Hepatitis B Virus (HBV)

The Demographic distribution of the subjects and incidence of Hepatitis B Virus (HBV) is presented in Table 1. A total of 115 subjects participated in the study all of which are diagnosed with hepatitis B virus. However, 92 subjects were confirmed positive for Hepatitis B Surface Antigen (HBsAg).

Table 1. *Demographic distribution of the subjects and incidence of Hepatitis B Virus (HBV) with percentage prevalence*

Parameters	Number	HBV+
Age		
10 - 20	15(13%)	11(12%)
21 – 30	44(38%)	38(42%)
31 – 40	19(17%)	14(15%)

41 – 50	21(18%)	17(18%)
51 - 60	16(14%)	12(13%)
Sex		
Male	70(61%)	57(62%)
Female	45(39%)	35(38%)
Maritalstatus		
Single	47(41) %	41(44%)
Married	49(43%)	42(46%)
Widow	8(07%)	3(3%)
Divorce	11(09%)	6(7%)
Educationstatus		
Primary	6(5%)	1(01%)
Secondary	41(36%)	34(37%)
Tertiary	40(35%)	33(36%)
Others	28(24%)	24(26)
Religion		
Islam	84(73%)	69(75%)
Christianity	31(27%)	23(25%)
Types of resident		
Urban	72(63%)	62(67%)
Semi-urban	30(26%)	24(26%)
Rural	13(11%)	6(07%)
Social status		
Student	41(36%)	30(33%)
Civil servant	21(18%)	17(18%)
Business	25(22%)	21(23%)
House wife	28(24%)	24(26%)

Evaluation of Hepatitis B Disease

The hepatitis B disease was evaluated using liver enzyme tests by detecting the level of Alanine Amino-transferase (ALT), Aspartate Amino-transferase (AST), Alkaline Phosphatase (ALP), and liver function tests (LFTs) that includeDirect Bilirubin (D_B) and Total Bilirubin(T_B) tests. Hepato cellular carcinoma has the highest occurrence with 42 subjects, followed by liver cirrhosis (20), asymptomatic hepatitis B (15) and acute hepatitis (10) while chronic hepatitis has the least incidence with 5 subjects.

Table 2. *Evaluation of hepatitis B condition among confirmed cases of hepatitis B cases*

Liver condition	Male subject	Female subject	Total	t value
Acute hepatitis	5(5.4%)	5(5.4%)	10(10.8%)	0.2369*
Asymptomatic HB	10(10.9%)	5(5.4%)	15(16.3%)	
Chronic hepatitis	2(02.2%)	3(3.3%)	5(05.5%)	
Liver cirrhosis	10(10.9%)	10(10.9%)	20(21.8%)	
Hepato-cellular carcinoma	30(32.6%)	12(13.0%)	42(45.6%)	
Total	**57(62%)**	**35(38%)**	**92(100%)**	

Key: *with calculated t value 0. 2369 less than the table value of 2.132 (4df), values between male and female subjects are considered not significantly different at $p<0.05$

Detection of Hepatitis D Virus (HDV)

The distribution of hepatitis D among hepatitis B subjects is presented in Table 3. Based on the results, the categories were based on age group. The result showed that 5 out of 92 subjects have hepatitis B and D co-infections. Age category 21-30 and 31-40 years have the highest co-infection with 2 subjects in each case while 41-50 years has 1 subject.

Table 3. *Detection of Hepatitis D virus among the confirmed 92 subjects with hepatitis B*

Age	Subjects	HBV+	HDV+	HDB-	t value
10 - 20	15(13%)	11(9.6%)	0(00%)	11(9.6%)	3.680*
21 – 30	44(38%)	38(33.0%)	2(2.2%)	36(30.8%)	
31 – 40	19(17%)	14(12.2%)	2(2.2%)	12(10.0%)	
41 – 50	21(18%)	17(14.8%)	1(1.0%)	16(13.8%)	
51 - 60	16(14%)	12(10.4%)	0(00%)	12(10.4%)	
Total	**115(100%)**	**92(80%)**	**5(5.4%)**	**87(74.6%)**	

Key :*with calculated t value 3.680 higher than the table value of 2.132 (4df), the prevalence of Hepatitis B and D co-infection among the subjects based on age categories are considered significantly different at $p<0.05$

Prevalence of Hepatitis D with Respect to Sex

The prevalence of Hepatitis D virus with respect to sex among Hepatitis B subjects is presented in Table 4. The result indicated that the hepatitis B and D co-infection is higher among female with 3 subjects accounted for 3.2% than male with 2 subjects (2.2%).

Table 4. *Prevalence of Hepatitis D virus with respect to sex*

Sex	Subjects	HBV+	HDV+	HDB-	t value
Male	70(61%)	57(49.6%)	2(2.2%)	55(47.4%)	0.9571*
Female	45(39%)	35(30.4%)	3(3.2%)	32(27.2%)	
Total	**115(100%)**	**92(80%)**	**5(5.4%)**	**87(74.6%)**	

Key: *with calculated t value 0.9571 less than the table value of 6.314 (1df), the prevalence of Hepatitis B and D between male and female subjects are considered not significantly different at $p<0.05$

HDV Antibody in HBsAg Related Liver Diseases

The prevalence of HDV antigen in the 92 positive HBsAg patients with different or various related liver diseases is presented in Table 5. HDV antigen did not appear in the positive HBsAg patients with Acute and Asymptomatic Hepatitis B Virus. 1 patient among the 5 patients with Chronic Hepatitis B Virus tested positive for HDV antibody (i.e HDV antigen percentage in chronic patients is 20%) and 1 patient with Liver Cirrhosis also tested positive for HDV antibody (i.e HDV antigen percentage in patients with liver cirrhosis is 5%). 3 patients among the 42 patients with Hepato-cellular Carcinoma (i.e Liver Cancer) tested positive for HDV antibody making 7.14% out of the 80% of patients with Liver Cancer.

Table 5. *HDV antibody in HBV related liver diseases*

Liver disease	Number (n)	HDV+	t value
Acute Hepatitis	10(11%)*	0(00%)	2.9224*
Asymptomatic HB	15(16%)*	0(00%)	
Chronic Hepatitis	5(05%)*	1(1.1%)	
Liver cirrhosis	20(22%)*	1(1.1%)	
Hepato cellular carcinoma	42(46%)*	3(3.2%)	
Grand Total	**92(100%)**	**5(5.4%)**	

Key: *with calculated t value 2.9224 higher than the table value of 2.132 (4df), there is significant different at $p<0.05$ on the prevalence of Hepatitis D virus based on related liver diseases.

Discussion

Worldwide, the pattern of hepatitis D infection is different from that of hepatitis B infection but have similar modes of transmission and it has been estimated that 15 million people with hepatitis B (HBsAg) are infected with hepatitis D [21]. Hepatitis B is a serious public health problem worldwide and one of the most common infectious disease globally[22]. It may occur as acute diseases of short duration, the liver damage. Symptoms of HBsAg usually appear from 40 days to 6 monthly after exposure. People with chronic hepatitis B who are infected with hepatitis D (co-infection) usually develop chronic (long-term) hepatitis D infection.A total of 115 patients with various forms of liver disease were screened for HBsAg. Ninety two of the subjects which accounted for 80% tested positive for HBsAg. These were made up of 10 patients with acute hepatitis (10.9%), 15 patients with asymptomatic infection (16.3%), 5 patients with chronic hepatitis (5.43%), 20 patients with liver cirrhosis (21.73%) and 42 patients with primary liver cell carcinoma (45.65%).

The disease spectrum of HBV infection can be grouped into early disease (acute hepatitis and asymptomatic infection) and late infection (chronic hepatitis, liver cirrhosis and hepato-cellular carcinoma). Advanced stages of HBV infection (liver cirrhosis and primary cell carcinoma) accounted for about 73% of the patients with HBV-related liver diseases who participated in this study. This is most likely because of late presentation which is a major problem in virtually all diseases encountered in developing countries. Even when patients are discovered at the stage of asymptomatic infection, follow up is usually a problem, because they often default and only reappear in hospital at a late stage when the chances of cure are almost nonexistent. This calls for concerted and sustained efforts at health education of the populace on the need for routine medical checks so that individuals with early stages of the disease can be identified, followed up and prompt interventions instituted as appropriate. The reason for the high rate of HBV in this study is that the enrolled patients were diagnosed to have chronic liver disease, of which HBV is the major cause. The reported rate for HBV infection in this study is very high as compared to a previous study by Ola and Olusanya [23] in Ibadan – Nigeria, which reported that 57.1% of patients with primary liver cell carcinoma were positive for HBsAg. In Jos and Gombe, prevalence rate showed 25.9% and 26.5% respectively among patients with human immune deficiency syndrome (HIV) [15,24]. According to the present study, Hepatitis B virus prevalence rate was higher in males (60.4%) than in females (39.6%), with the majority of the subjects in the age range of 26 – 45 years old. This could be due to the fact that this age group is mostly exposed to multiple risk factors associated with HBV.

The prevalence of anti-HDV in this study was 5.43%. Furthermore, there have been reports of regional variability in HDV prevalence in some parts of Africa and this may also be operative in Nigerian patients. Anti-HDV was demonstrated in 5 patients (5.43%), made up of 2 males and 3 females. The gender specific prevalence of anti-HDV was 13.0% for males and 10.5% for females. This is finding was in conformity with that of Andrade et al., [25]in the Western Brazilian Amazon where the prevalence is significantly higher in males than female subjects. Table 5illustrates the anti-HDV sero-positivity in the different forms of liver diseases. In summary, the prevalence of anti-HDV was 0% in acute hepatitis, 0% in asymptomatic infection, 0% in chronic hepatitis, 5.0% in liver cirrhosis and 7.14% in hepato-cellular carcinoma.

The prevalence of anti-HDV in patients with late stages of HBV-related liver diseases (chronic hepatitis, liver cirrhosis and hepatocellular carcinoma) was 15.0% compared to a lower prevalence of 4.3% in patients with early infection (chronic hepatitis, Liver cirrhosis and Hepatocellular carcinoma infection). Even though the difference was not statistically significant, this might be an important observation because it suggests that the infection has become a super-infection. This result supported that of Nwokediuko and Ijeoma [16]who reported that in Nigeria patients with acute hepatitis and asymptomatic infection has prevalence of 4.3% while in patients with chronic hepatitis, liver cirrhosis and primary liver cell carcinoma has prevalence of 15%.

Conclusion

The results from this study have demonstrated the emergence of HDV co-infection HBV in Aminu Kano Teaching Hospital a with lower prevalence rate of 5.43%.The females with Chronic HBV infection have high percentage prevalence of HDV co-infection than the males. In the study, we observed that all HDV

infected patients were positive for HBV and thus this shows that HBV, HDV have a common mode of transmission and both have the same serological markers as HDV cannot persist without the presence of HBV. However, the percentage prevalence of HBV infection was observed to be high with the males having the higher percentage of HBV than their female counterparts. It is recommended that public enlightment and knowledge of HBV and HDV infections transmission should be provided to people in order to reduce the risk of being infected with both diseases.

Acknowledgement

The authors wish to acknowledge to the staff of Aminu Kano Teaching Hospital for their support and samples provision. Thanks to Associate Professor Ismail Ahmad (KUST Wudil).

References

[1] World-Health-Organization (WHO). Hepatitis B, Fact sheet No 204, updated 2015.

[2] Hadziyannis SJ. Review: hepatitis delta. J Gastroenterol Hepatol; 1997; 12: 289–98.

[3] Andrade ZA, Lesborders JL and Ravisse P. Fulminate hepatitis with microvescicular steatosis (a histological comparison of cases occurring in Brazil-Labrea hepatitis- and in central Africa-Bangui hepatitis). Rev. Soc. Bras. Med. Trop.1992; 25,155–160.

[4] Wedemeyer H. Re-emerging interest in hepatitis delta: new insights into the dynamic interplay between HBV and HDV. J Hepatol; 2010; 52:627–29.

[5] World-Health-Organization (WHO). Hepatitis Delta Virus. Global Alert and Response (GAR). http://www.who.int/csr/disease/hepatitis/whocdscs. 2011

[6] Taylor JM. "Hepatitis delta virus". Virology. 2006; 344 (1): 71-76. PMID 16364738. doi:10.1016/j.virol.2005.09.033.

[7] Modahl LE and Lai MM. The large delta antigen of hepatitis delta virus potently inhibits genomic but not antigenomic RNA synthesis: a mechanism enabling initiation of viral replication. J Virol.2000;74:7375–80.

[8] Alvarado-Mora MV, Locarnini S, Rizzetto M and Pinho JR. An update on HDV: virology, pathogenesis and treatment. Antivir Ther. 2013; 18:541–8.

[9] Radjef N, Gordien E, Ivaniushina V, Gault E, Ana€is P and Drugan T.et al.(2004) Molecular phylogenetic analyses indicate a wide and ancient radiation of African hepatitis Delta virus, suggesting a Deltavirus genus of at least seven major clades. J Virol 2004;78:2537-2544.

[10] RizzettoM.Hepatitis D: thirty years after. J. Hepatol.50,1043–1050. Provides an overview of HDV virology, epidemiology and unsolved problems on HDV-related liver disease. 2009

[11] Makuwa M, Mintsa-Ndong A, Souquiere S, Nkoghe D, Leroy EM and Kazanji M. Prevalence and molecular diversity of hepatitis B virus and hepatitis delta virus in urban and rural populations in northern Gabon in central Africa. J Clin Microbiol.2009; 47:2265–8.

[12] HughesSA, Wedemeyer H and Harrison PM. Hepatitis delta virus. Lancet. 2011;378:73–85.

[13] Cakaloglu Y, Akyuz F, Bozaci M, Ibrisim D, Pinarbasi B and Demir K et al. Prevalence and clinical significance of SEN-H virus in chronic hepatitis B, C and delta infections in Turkey. Turk J Gastroenterol. 2008; 19:104–8.

[14] Nakasone H, Sakugawa H, Shokita H, Nakayoshi T, Kawakami Y and Kinjo F. et al. Prevalence and clinical features of hepatitis delta virus infection in the Miyako Islands, Okinawa, Japan. J Gastroenterol. 1998; 33:850–4.

[15] Ojo OSand Akonai A, Thursz M, Ndububa DA, Durosinmi MA and Adeodu OOet al. Hepatitis D virus antigen in HBsAg positive chronic liver disease in Nigeria. East Afr Med J.1998;75:329–31.

[16] Nwokediuko SC and Ijeoma U. Seroprevalence of antibody to HDV in Nigerians with hepatitis B virus-related liver diseases. Niger J Clin Pract.2009;12:439–42.

[17] National Population Commission (NPC). National population census result, 2006 Abuja Nigeria

[18] Nwabuisi EC and Onile BA "Male Infertility among Sexually Transmitted Disease Clinic Attendees in Ilorin," Nigerian Journal of Medicine, Vol. 10, No. 2, pp. 68-71. Obstetrique and Fertilite 2001; 33(9): 691 – 697

[19] Djebbi A, Rebai WK, Bahri O, Hogga N, Sadraoui A and Triki H. Serological markers, viral RNA and genotype of hepatitis delta virus in HBs antigen positive Tunisian patients. Pathol. Biol. 2009; 57,518–523.

[20] Changotra H, Dwivedi A, Nayyar AK and Sehajpal PK. Diagnosing different stages of Hepatitis B infection using a competitive polymerise chain reaction assay. Indian Journal of Medical Microbiology 2008; 26(2): 138-142

[21] Farci PF. Treatment of chronic hepatitis D: New advances, old challenges Hepatology. 2006; 44(3): 536-39.

[22] Locarnini S. "Molecular Virology of Hepatitis B Virus". Seminars in Liver Disease. 2004; 24: 3–10. PMID 15192795. doi:10.1055/s-2004-828672.

[23] Ola SO, Jaiyesimi AE and Olusanya OO. Coinfection of HIV and HBV among Nigerians. J Med Virol. 2005;83:710–6.

[24] Mustapha SK and Jibrin YB. The prevalence of hepatitis b surface antigenemia in patients with human immunodeficiency virus infection in Gombe, Nigeria. Ann Afrii Med. 2004; 4:10-15

[25] Andrade ZA, Lesborders JL and Ravisse P . Fulminate hepatitis with microvescicular steatosis (a histological comparison of cases occurring in Brazil-Labrea hepatitis- and in central Africa-Bangui hepatitis). Rev. Soc. Bras. Med. Trop. 1992; 25,155–160

Citation: **Waiya S.A, Taura, D.W, Shehu A. A, Yahaya S.M, Ali M, Garba M.** *Prevalence of Hepatitis D Virus Antigens Among Sero-Positive Hepatitis B Surface Antigen (HBsAg) Patients Attending Aminu Kano Teaching Hospital (AKTH), Kano. Archives of Immunology and Allergy. 2018; 1(2): 67-74.*