Seroprevalence of Hepatitis C Virus Infection among Patients Living with Human Immunodeficiency Virus Attending Aminu Kano Teaching Hospital, Kano, Nigeria

Hassan, M. S., Dutsinma U. A[1*]., Ramadan, T., Akande, A. O., Mohammed, Y., Usman, J. N., Babayo, A., Idris, A. M., Abdullahi, A. I., Ilah, I. N., Safiyanu, M. B., Dayyab, D. and Umar, A. A[2].

Department of Medical Microbiology and Parasitology, Faculty of Clinical Sciences, Bayero University, Kano, Nigeria

[1]Department of Microbiology, Faculty of Life Sciences, Bayero University, Kano, Nigeria

[2]Department of Community Medicine, Faculty of Clinical Science, College of Health Science, Bayero University,

Kano, Nigeria

*Corresponding author: Tel: +2348035892096; amikibiya@gmail.com, ualiyu@gmail.com.

Abstract: Hepatitis C virus (HCV) is a major disease burden on the world and man is the only known natural host of HCV. HCV infection depends on age, sex, and immune-competence at the time of infection. In most immuno-competent adults, 75% to 85% develop chronic HCV infection. Human immunodeficiency virus (HIV) increases the pathological effect of HCV infection and potentiates the re-activation of latent hepatitis infections due tolowered immunity. About 10% of HIV-positive individuals are HCV antibody carriers. The present study aimed at determines the HCV/HIV co-infection among patients attending Antiretroviral clinic of Aminu Kano Teaching Hospital, Kano, Nigeria. One hundred and eighty (180)known HIV-positive are screened for the presence of HCV infection using HCV antibody Enzyme-Linked Immunosorbent Assay (ELISA) kit according to the manufacturer's instructions for qualitative detection in plasma. Of the 180 subject screened for HCV, an overall prevalence of 5 (2.8%) were found. Subject aged 41 – 50 years had the highest seroprevalence (5.6%), followed by those aged 0 – 20 years (4.4%) and least seroprevalence was among those aged 21 – 30 and >50 years (0.0%). The highest seroprevalence was obtained among the subject with CD4 cell count of 0 – 200cell/mm3 and those on antiretroviral therapy for about 1 – 5 years. The finding of this study suggested that all HIV-positive should be routinely screened for HCV since about 10% of HIV-positive are HCV carriers and a decline in CD4+ cell counts will increase the chance of developing chronic HCV infection.

Keywords: Hepatitis C virus, HIV-positive, ELISA, Co-infection, Liver

INTRODUCTION

Hepatitis C is an infectious disease caused by the hepatitis C virus (HCV) that primarily affects the liver. An estimated 130 – 200 million people are infected with the Hepatitis C virus worldwide and occur most commonly in Africa, Central, and East Asia (CDC, 2015). About 1.75 million new cases were estimated worldwide in 2015 (WHO, 2017). In Nigeria, the prevalence rate of HCV based on previous studies varies between 2.3% – 5.7% (Nwannadi *et al*., 2012; Okerentugha *et al*., 2015). Despite this high prevalence, prior research has shown that much younger than 30 years of age are unaware of their status (Korthuis *et al*., 2012). Hepatitis C virus causes death due to liver disease worldwide among HIV positive individuals. HIV increases the pathological effect of hepatitis viruses and potentiates the re-activation of latent hepatitis infections due to lowered immunity (Pamela *et al*., 2016). Hepatitis C virus infection depends on age, sex, and immune status at the time of infection. In most immune-competent individual, 75% - 85% develop chronic HCV infection. Chronic infection may result in a healthy carrier state, liver cirrhosis, and hepatocellular carcinoma. For individuals who develop acute liver failure about 80% die within days or weeks after infection. There is a high chance of transmission to the newborn from a highly infectious mother and 30% of children below 20 years develop chronic HCV infection (Sungkanuparph *et al*., 2004; Alter, 2006). About 10% of HIV-positive individuals are HCV antibody carriers (Massroor *et al*., 2007). HIV disease progression is the presence of co-morbidities and opportunistic infections (Ajegena *et al*., 2017).

Human immunodeficiency virus (HIV) and HCV are RNA viruses that have similar modes of transmission and hence co-infections are common and potentiate each other (Benhamou, 2004; Soriano *et al*., 2006). AlsoHIV increases the risk of re-activation of previously existing asymptomatic and chronic HCV infections. HCV/HIV co-infected individuals have a threefold chance of getting hepatotoxicity (Sulkowski, 2007). Therefore, the proper diagnosis of HCV among HIV-positive individuals is important and facilitates better management of patients (Soriano *et al*., 2006). The present study aimed to determine the HCV/HIV co-infection among patients attending antiretroviral clinic of Aminu Kano Teaching Hospital, Kano, Nigeria.

MATERIALS AND METHODS

Study Area

The study was conducted at Aminu Kano Teaching Hospital located in the Kano metropolis. Kano State is located in the Northwest geopolitical zone of Nigeria. It comprises 44 Local Government Area with an estimated population of over 13 million and 20,760 km^2. It lies between latitudes 10^o 33N to 11^o 15N and longitudes 34^oCE to 8^o 20CE (NBS, 2018).

Study Population

The study populations were HIV positive patients attending Antiretroviral Therapy Clinic at Aminu Kano Teaching Hospital, Kano State.

Study Design

This study was a cross-sectional study to see HIV positive patients for the detection that pften detects Hepatitis C virus (HCV), which may cause liver damage malfuction.

Inclusion and Exclusion Criteria

Only HIV-positive patients who had been receiving antiretroviral therapy were included in this study while HIV negative and non-consulting HIV positive patients were not included in the study.

Ethical Approval

Ethical approval was obtained from Aminu Kano Teaching Hospital ethical and research committee before the commencement of the study with reference number (AKTH/MAC/SUB/12A/P- 3/VI/1837).

Consent of Subjects

A consent form containing the research topic, the researcher's name, and the purpose of the study was administered to the patients for their consent. Only patients who consented to participate were included in the study.

Sample Collection and Processing

The subjectsrandom sampling technique obtained the subjects for the study. Patient information was collected using structured questionnaires, including patient identification number, age, sex, clinical data, and other socio-demographic characteristics. The samples were collected for a period of two months from February to April 2019. A blood sample (5ml) was collected aseptically from each of the subjects described by Cheesebrough, (2000). Samples collected were dispensed into sterile containers and was allowed to clot and retract. The serum was separated by centrifugation at 3500rmps for five minutes to avoid haemolysis of red blood cells. The serum separated was transferred aseptically into a plain sterile container and stored at -200C until needed for assay (Junaid *et al*., 2014).

Serological Test for HCV

All samples were analyzed using HCV antibody Enzyme-Linked Immunosorbent Assay (ELISA) test system (Dia. Pro. Diagnostics Bioprobes, Italy). All assay protocols were done according to the manufacturer’s instructions.

Assay Procedures

The plasma from every participant was diluted with 200µl of DILSPE. Each sample was further diluted with 50µl of DILAS alongside the negative controls in triplicate, the calibrator in duplicate, and a positive control provided by the kit manufacturer. After the microplate was incubated for 45mins at +37oC and the wells washed, all the wells were then treated with 100µl of the enzyme conjugate except the first blanking well.

The microplate was incubated again for another 45mins at the same temperature and the chromogen/substrate mixture was added after the second washing and incubated for 15mins at room temperature. The reactions were stopped with 100µl of sulphuric acid and the optical density (O.D) read at 450 nm immediately. The cutoff value for each batch was determined and individual results were interpreted as Negative (<0.9) and positive (>1.1) and equivocal (0.9-1.1).

Data Analysis

All the data generated were collated, checked, and entered into a database design using M.S excel spreadsheet and analyzed using Statistical Package for Social Science (SPSS) Version 25.0. The values were expressed as means and percentages. The Chi-square test determined a comparison of variable. The level of significance of $P<0.05$ was employed.

RESULTS

Out of a total of 180 HIV-positive patients, 5 (2.8%) were HCV seropositive as shown in figure 1.

Out of 180 HIV infected patients, 83 (46.1%) were males and 97 (53.9%) were females. Of the 83 males, HIV/HCV Co-infection was observed in 3 (3.6%) while 2 (2.1%) females were also Co-infected with HIV/HCV out of a total of 97 female patients (Table 1). Among all the 180 HIV patients examined, the highest prevalence of HCV Co-infection was observed among patients in the age group 41 – 50 years 2 (5.6%), followed by those aged 31 – 40 with 2 (3.7%). Patients aged 21 – 30 and >50 years had the least prevalence rate of HIV/HCV co-infection (Table 1).

The distribution of HCV seropositive with patients' clinical history shows the highest incidence of HIV/HCV co-infection among patients with low CD4 of 0 – 200cell/mm3 and null incidence rate at a high CD4 count of ≥500cell/mm3. Based on ART duration most subjects with HCV co-infection are at ART for 1 – 5 years (Table 2).

Table 3 shows the prevalence of HCV co-infection in relation to possible risk factors for acquiring HCV infection. Patients sharing sharp objects show greater incidence (8.3%) of HCV co-infection followed by those with a history of jaundice (8.0%) and scarification (4.3%) respectively. No patients were found to have HCV co-infection among patients with a history of blood transfusion and surgery.

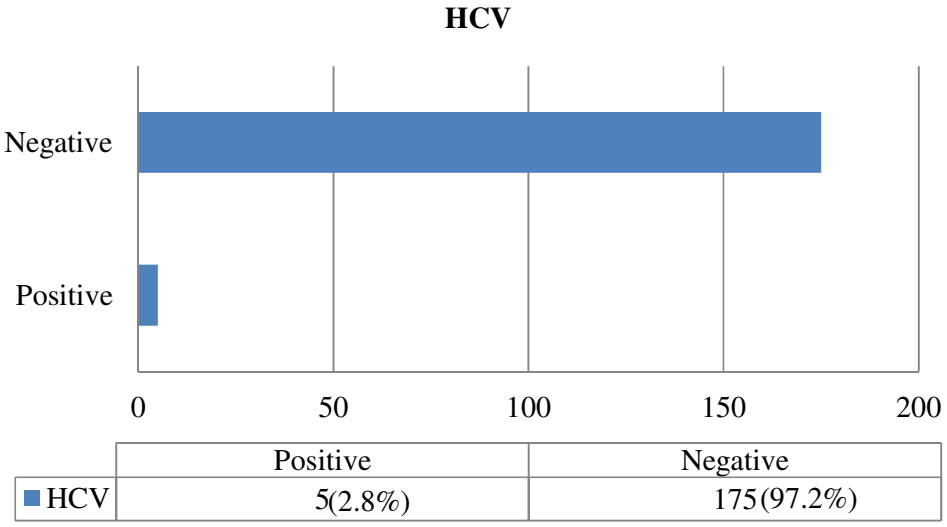

Figure 1: Showing the Prevalence of HCV infection among HIV positive patients attending antiretroviral therapy clinic in AKTH

Table 1: Prevalence of HCV Infection With respect To Demographical parameters

Demographic Parameters	No. Examined (%)	HCV	
		No. positive (%)	No. Negative (%)
Age Group (Years)			
0 – 20	23 (12.78)	1 (4.4)	22 (95.6)
21 – 30	55 (30.56)	0 (0.0)	55 (100.0)
31 – 30	54 (30.00)	2 (3.7)	52 (96.3)
41 – 50	36 (20.00)	2 (5.6)	34 (94.4)
>50	12 (6.67)	0 (0.0)	12 (100.0)
Gender			
Male	83 (46.11)	3(3.6)	80(96.4)
Female	97 (53.89)	2(2.1)	95(97.9)
Total	**180 (100.00)**	**5 (2.8)**	**175 (97.2)**

Table 2: Prevalence of HCV In Relation To Clinical History of the Study Subjects

Clinical History	No. Examined (%)	HCV	
		No. positive (%)	No. Negative (%)
CD4 Cell Count			
0 – 200	66	3 (4.6)	63(95.5)
201 – 499	78	2 (2.6)	76(97.4)
≥500	36	0 (0.0)	36(100.0)
Duration on ART (Year)			
<1	76	1 (1.3)	75(98.7)
1 – 5	24	3 (12.5)	23(95.8)
>5	80	1 (1.3)	79(98.8)
Total	**180**	**5(2.8)**	**175(97.2)**

Table 3: Prevalence of HCV in Relation to Possible Risk Factors for Acquiring HCV Infection

		HCV		*P-value*
Risk Factors	**No. Examined (%)**	**No. positive (%)**	**No. Negative (%)**	
Blood Transfusion				
Yes	18	0 (0.0)	18 (100.00)	0.000
No	162	5 (3.1)	157 (96.9)	
Surgery				
Yes	16	0 (0.0)	16 (100.0)	0.9999
No	164	5 (3.0)	159 (97.0)	
Sharing of Sharp Object				
Yes	12	1 (8.3)	11 (91.7)	0.5892
No	168	4 (2.4)	164 (97.6)	
History of Jaundice				
Yes	25	2 (8.0)	23 (92.0)	0.2849
No	155	3 (1.9)	152 (98.1)	
Scarification				
Yes	47	2 (4.3)	45 (95.8)	0.7817
No	133	3 (2.3)	130 (98.5)	

DISCUSSION

In this study, the overall seroprevalance of 2.78% for HCV among HIV-positive individuals was got, comparable to the 2.6% prevalence obtained in previous studies conducted in Rwanda (Mutagoma *et al*., 2017) and 3% in Port Harcourt Nigeria (Okerentugha*et al*., 2015). This may be due to the similarities of the study subjects.It is also slightly higher than the prevalence of 2.3% in Abuja, Nigeria (Adewale *et al*., 2009), 1.69% in India (Raizada *et al*., 2011), and 1% in Nairobi Kenya (Harania *et al*., 2008). The prevalence obtained in this study is lower than the rate of HCV infection recorded in Ghana 7.7% (Ephraim *et al*., 2015), Egypt 6.1% (Zenebe *et al*., 2015) and elsewhere in Nigeria; 4.5% in Benin (Nwannadi *et al*., 2012) and 5.7% in Jos (Udeze *et al*., 2011). This may be due to differences in geographical variation, cultural practices, sexual behaviors, nonadherence to drug administration, and laboratory procedures employed for HCV detection.

The highest prevalence (5.6%) of HCV was found among the age groups of 41-50years which agrees with a similar study by Chiekulle *et al.* (2013). This may be becouse HIV deals with the age and immunocompetence of individuals at a time of infection.The prevalence obtained was higher among males (3.6%) than females (2.1%). This result is consistent with a previous study by Narayanasamy *et al.* (2016) and contrary to a study by Taiwo *et al.* (2012). The higher prevalence among men could be related to the fact that men are more exposed to HCV risk factors such as intravenous drug injection, sharp objects, and alcohol consumption.

The outcome of this study shows that HCV and HIV co-infected subjects had a high prevalence (4.5%) of CD4+ cell counts that from range 0 – 200cells/µl which is indicating severe immunosuppression among this group. Studies by Olawumi *et al.* (2014) had found that HIV and HCV co-infection was associated with a decline in CD4+ cell count. It is also similar to a previous study by D'almeida *et al*. (2017) in Cotonou. These may be attributedbecause CD4+ cell count is the most used clinical selection criterion to determine ART eligibility for HIV-infected individuals (D'almeida *et al*., 2017). Earlier studies revealed that co-infection between the hepatitis C virus and HIV has been associated with a rapid decline in the CD4+ count, rapid progression of HIV infection, and increased morbidity and mortality (Thomas, 2012).

All the subjects in this study were on ART. However, the duration of ART has no significant impact on the rate of HCV infection which is contrary to a previous study in Cotonou (D'almeida *et al*., 2017) that shows an association between HCV infection and length of ART. This study shows a significant impact between socio-demographic risk factors and HCV which is similar to a previous study by Bala *et al.* (2012) and at variance with a study in Istanbul, Turkey (Oziem *et al*., 2014). It is also contradictory to previous findings of Sheyin *et al.* (2011) who reported that in comparison with other risk factors, blood or blood products transfusion was found to be the highest risk factor for acquiring HCV infection. This may be due to a lack of adequate knowledge on HCV, how it can be transmitted, and feeble efforts to manage HIV patients.

CONCLUSION

In conclusion, this study shows a high prevalence (2.8%) of HCV among HIV positive patients in the study area. The study also revealed that males have a more prevalent rate of HCV infection than females were adults' ≥40years have the highest prevalence. The outcome of this study shows that there is a decline in CD4+ cell counts (≤200cells/µl) among these patients co-infected with hepatitis C virus and HIV. The study indicated that socio-demographic risk factors likely to be associated with HIV/HCV co-infection may include blood

transfusion, history of jaundice, body scarification, settlements, educational background. The study recommends that HIV positive patients should be routinely screened for HCV before initiation of highly active antiretroviral therapy as this practice would guide the correct choice of the drug combination. Government should organize public enlightenment programs on the effects of HCV and how it can be transmitted especially among the risk groups and there is need for continuous search for an HCV vaccine to reduce the burden of the infection on the risk group.

REFERENCES

Abiodun, C.J., Bolayi, O.O., and Sebastine , O.O. (2014). Prevalence of Hepatitis C virus Antibody Among under graduates in Ogbomosho southwestern Nigerian. *African Journal of Infectious Disease,* 8 (2):40 – 45.

Adewole, O. O., Anteyi, E., Ajuwon, Z., and Wada, I. (2009). Hepatitis B and C Virus Coinfection in Nigerian Patients with HIV Infection. *Journal of Infections in Developing Countries,* 3(5):1221 – 1222.

Ajegena, S.A., Oti, B.V., Pennap, R.G., Richard, M. (2017). Prevalence of HBsAg and HBV serotypes using antigen detection and PCR methods among human immunodeficiency virus patients accessing healthcare in a Tertiary Healthcare Facility in central Nigeria. *Journal of Advance Microbiology*, 3(3): 1-10.

Alter Miriam, J. (2006). Epidemiology of viral Hepatitis and HIV Co-infection. *Journal ofHepatology*, 44(1):6–9.

Bala, J.A., Kawo, A.H., Mukhtar, M.D., Sarki, A., Magaji, N., Aliyu I.A. and Sani, M.N. (2012). Prevalence of hepatitis C virus infection among blood donors in some selected hospitals in Kano, Nigeria.*International Research Journal of Microbiology,* 3(6):217-222.

Benhamou, Yves. (2014). Antiretroviral therapy and HIV/Hepatitis B virus co infection. *Journal ofClinical Infectious Diseases*,38(2):98–103.

Centers for Disease Control and Prevention CDC., (2016).*Hepatitis C FAQs for Health Professionals. Archived from the original on 21 January 2016.* Retrieved 4 February 2016.

Cheesebrough, M. (2000). District laboratory practice in Tropical countries. Part 2. *Cambridge University Press UK,* Page 279-282.

Chiekulle, K.D., Emmanuel, C.O., Oguamanam, O.E., Jerome, E.A., and Nathan, C.N. (2013). Seroprevalence of Hepatitis B and C Virus among HIV Patients in a Suburban University Teaching Hospital in South-East Nigeria. *Pan African Medical Journal,* 16-17.

D'almeida, M., Guidibi-Zohoun, L., Adedemy, D.J., Lalya, H.F., and Gbogbo, H. (2017). Prevalence and Factors Associated with HIV and Hepatitis B and Hepatitis C Co-Infection in Children Attended at the Hubert KoutoukouMaga National University Teaching Hospital of Cotonou. *Pediatric Therapy,* 7: 330.

Ephraim, R., Donko, I., Sakyi, S.A., Ampong, J.,andAgbodjakly, H. (2015). Seroprevalence and Risk Factors of Hepatitis B and C Infection among Pregnant Women in the Ashanti Akim North Municipality of the Ashanti Region Ghana a cross sectional study. *Journal of African HealthScience*, 15:709 –713.

Harania, R.S., Karuru, J., Nelson, M., and Stebbing, J. (2008). HIV, Hepatitis B and C coinfection in Kenya. *AIDS*, 22(10):122 –132.

Korthuis, P.T., Feaster, D.J., Gomez, Z.L., Das, M., and Tross, S. (2012). Injection behaviors among

injection drug users in treatment: the role of hepatitis C awareness. *Addict Behavoir,* 37: 552-555.

Massroor, A.M., Zahoor, S.Z., Akbar, S.M., Shaukat, S., Butt, A.J., Naeem, A., Sharif, S., and Angez, M. (2007). Molecular epidemiology of Hepatitis B virus genotypes in Pakistan. *BMC Infectious Diseases*, 7:115.

Mutagoma, M., Balisanga, H., Sebuhoro, D., Mbituyumuremyi, A., Remora, E., and Malamba, S.S. (2017). Hepatitis C Virus and HIV coinfection among pregnant women in Rwanda. *Biomedical Journal Infectious Disease*,17:167.

Narayanasamy, K., Jasmine, J.J., and Ganesh, M.J., (2016). Current Prevalence of HCV in Patients with Liver Disease and its Related Profile. *Colorectal Cancer,* 2:4.

National Bureau of Statistics (NBS) (2018). The latest population figures from National Bureau of Statistics you need to see; Business Insider by Pulse.

Nwannadi, I.A., Adao, O.O., Bazauye, G.N., Omoti, C.E., and Halim, N.K. (2012). Seroprevalence of Hepatitis C virus Antibody in sickle cell Aneamia patients in Benin city Nigeria. *Gomal Journal of Medical Sciences*, 10:11.

Okerentugba, P.O., Uchendu, S.C., and okonko, I.O. (2015). Prevalence of HIV Among pregnant Woman in Rumubiakani, Port Harcourt, Nigeria. *Public Health Journal,* 5:58 -65.

Olawumi, H.O., Olanrewaju, D.O., Shittu, A.O., Durotoye, I.A., and Akande, A.A. (2014). Effect of hepatitis-b virus co-infection on CD4 cell count and liver function of HIV-infected patients. *Ghana Medical Journal*, 48: 96-100.

Oziem, A.A., Mulahit, Y., Hayat, K.K., Fatma, S., Alper, G., Bahadir, C., Bugul, M., Nail, O., Dilek, Y,S., Resat, O., and Fehmi, T. (2014). Low Prevalence of HCV Infection among HIV Positive Patients. Data from a large scale cohort study in Istanbul, Turkey. *International Monthly Journal of Hepatology*, 14(8); 18128.

Pamela Valva, Daniela A. Ríos, Elena De Matteo, and Maria V. Preciado, (2016). Chronic hepatitis C virus infection: Serum biomarkers in predicting liver damage; *Journals of Gastroentrology,*22(4): 1367–1381.

Raizada, A., Dwivedi, S.B., and Hattachary, S. (2011). Hepatitis B, Hepatitis C and HIV Coinfection at an Antiretroviral Centre in Delhi. *Tropical Journals,* 41 (3); 154 – 156.

Sheyin, Z., Jatau, E.D., Mamman, A.I, Randawa, A.J., Bigwan, I.E 2012. Detection of Hepatitis C virus among pregnant women in Kaduna State, Nigeria. *Wudpecker, Journal of Medical Science*,1(2): 012- 015.

Soriano, V., Barreiro, P., and Nuñez, M. (2006). Management of chronic hepatitis B and C in HIV coinfected patients. *Journal of Antimicrobial Therapy*, 57(5):815–818.

Sulkowski, M.S. (2007). Therapeutic issues in HIV/HCV co-infected patients. 600 North Wolfe Street, 1830 Building, Room 448, Baltimore, USA: *Viral Hepatitis Center, Johns Hopkins University School of Medicine,* MD 21287-0003.

Sungkanuparph, S., Vibhagool, A., Manosuthi, W., Kiertiburanakul, S., Atamasirikul, K., Aumkhyan, A., and Thakkinstian, A. (2004). Prevalence of Hepatitis B Virus and Hepatitis C Virus Co- infection with HIV in Thai Patients, International Conference for AIDS. *Journal of Medical Association, Thailand*, 87(11):1349–1354.

Taiwo, M.B., Samuel, E., and Emmanuel, F.O. (2012). HIV, Hepatitis B and C Viruses Coinfection among Patients

in a Nigerian Tertiary Hospital. *Pan African Medical Journal;* 12-100.

Thomas, D.L. (2012). Hepatitis C and human immunodeficiency virus infection. *Journal of Hepatology*, 36(5):201–209.

Udeze, A.O., Bamidele, R.A., Okonko, I.O., and Sule, W.F. (2011). Hepatitis C Virus Antibody Detection among First Year Students of University of Illorin, Nigeria. *World Journal of Medical Sciences,* 6: 162-167.

World Health Organization. (2017). *Hepatitis C, Fact Sheet*; WHO: Geneva, Switzerland.

Zenebe, Y., Muly, W., Yimer, M., and Abera, B. (2015). Seroprevalence and Risk Factors of Hepatitis C Virus Infection among Pregnant Women in Bahir Dar city, Northwest Ethiopia: cross sectional study. *Pan African Medical Journal,* 21:158-160.