Original Research Article

Clinico-Pathological Characteristics of HCV coinfection in Chronic HBV Liver Diseases Patients in North-central Nigeria

Abstract

Introduction: Viral hepatitis(HBV,HCV) coinfection worsen patient diagnosis and management approach. Local clinical and pathological data provides a basic diagnostic information necessary for patient management. Hence, this study evaluated theclinico-pathological characteristics of HCV coinfection in chronic HBV liver disease patients.**Methodology**: A total of 100 chronic HBV patients were enrolled into the study, with detailed demographic and clinical information documented into study database. Serodetection of HBsAg was determined by rapid immunochromatographic kit, and Anti-HCV by Enzyme linked immunosubent assay(ELISA)method. Heamatological indices analysedusing Systmexheamatologicalanalyser and biochemical profile, liver function tests by standard methods. Appropriate statistical method applied in analysis of study data.**Results**: The 100 study patients comprised of 65% males and 35% females, male to female ratio of 1.9:1, mean age of coinfected patient was 42.0±8.8years,with majority within theage-group of 41-50years. Anti-HCV seroprevalence of 10%, 7(10.8%) males and 3(8.6%) females, M:F 1:2.3.Histological diagnosis of liver biopsies,92% cases identified as chronic liver disease and 4% each as cirrhosis and hepatocellular carcinoma respectively. Anti-HCV detection in coinfected liver biopsies, 75% as hepatocellular carcinoma(HCC) cases, 25% as cirrhosis and 6.6% as chronic hepatitis. Significant association was observed between severity of liver disease and degree

of infection(p=0.01). Histological activity index score was significantly low in coinfectedpatients(5.2+4.2) compared with monoinfection(8.2+4.2)(p=0.01). There was siginificant difference in the biochemical and heamtological indicescoinfected and monoinfected patients(<0.05).**Conclusion:**The coinfection prevalence of 10% and the clinic-pathological picture was at variance with other Nigerian studies. The main findings revealed concomitant contribution of both hepatic viruses to development of chronic liver disease

Keywords, HCV, HBV, coinfection, chronic liver disease, north central Nigeria

Introduction

Globally, Hepatitis B and C remains a public health problem, with high prevalence and burden in sub-Saharan Africa and Asia responsible for chronic liver diseases, cirrhosis and hepatocellular carcinoma with high morbidly and mortality rate[1]. Characteristically, both hepatotropic viruses shared same mode of transmission although HBV infection predominantly acquired early in life via vertical and perinatal transmission while, HCV infection by contact with contaminated body fluid. According to the World Health Organisation, 350million and 170million are estimated to beinfected by HBV and HCV worldwide respectively, while approximatively 1 million died annually due to liver related cases including cancer[2,3]. Clinical implication and outlook of coinfection of HBV and HCV is complex, but common in viral hepatitis endemic region like sub-Saharan Africa, particularly West African region and Asia due to the shared mode of transmission[4].The exact prevalence is not known, it is projected to bebetween 1and 15% globally[5], 2-10% among HCV antibody positive patient and 3-20% among hepatitis B surface antigen carriers[6,7].

In Nigeria, national surveillance study of HBV infection is reported to be 12.2% [8] and Anti-HCV prevalence from pooled studies ranged between 0.4 and 5.5%[9], although these data varied with geographical location and predisposing risk factors. As epidemiological data of coinfection continued to evolve, few studies had reported coinfection prevalence in chronic liver disease subtypes, 20% in Maiduguri[10], 12.2% in Lagos[11], 11.8% in Jos[12]. Coinfection is common among IDU, heamodialysis patients, organ transplantation patient, HIV patents -positive patient, and beta-thalassemia patient10%[7]. Risk factors are more likely to occur among individuals aged over 40years, geographical variation, history of blood transfusion,, intravenous drug user, unsafe sex, history of genetic hematological disease[7].

Serological screening for HbsAg and Anti-HCV are routinely used for screening and early diagnosis of suspected liver disease cases while PCR assay of HBV-DNA and HCV-RNA are used as the precise marker of active HBV replication, possible variant infection and reliable marker of infectivity. Histological diagnosis of liver biopsy is used as the investigation of choice for assessment of inflammation and fibrosis that provides insight into assessment of liver damage, necro inflammatory changes and antiviral therapy. In addition, the Histological active Index(HAI) is used to grades/and score of hepatic disease development and progression of liver diseases. Of the histological findings, documented studies had reported conflicting changes in the liver diseases of coinfection and monoinfectionbased on the severity of liver diseases, while other studies reported no difference in the histological changes [13,14].However, some studies have reported necro inflammatory changes and fibrosis in coinfection[15]. The biochemical andheamatological tests are routinely used to complement the histological findings in the evaluation of disease progression. Based on this

observation, the study was conducted to evaluate the clinico-pathological profile of HCV coinfection of chronic liver disease cases.

Patients and Methods

The descriptive, cross-sectional study was conducted at the Gastroenterology Unit, Jos University Teaching Hospital between March and December 2005. The 600 beds capacity hospital, provides multimedicalspecialities to Nigerians and foreigners. The study protocol was approved by Jos University Teaching Hospital Institution review board. A well-structured study questionnaire and informed consent was administered to the study participants, which entails the following information, demographic, clinical data of past medical history, previous history of jaundice, blood transfusion, use of intravenous drugs and traditional surgery such as scarification, uvulectomy , tatto and alcohol intake

The inclusion criteria, patients who were positiveHBsAg for over six months and the following tests, full blood count, liver function test, HIV screening , prothrombin, abdominal ultrasound scan and liver biopsy. Exclusion criteria includes, HIV infection, contraindication to liver biopsy, pregnancy.

Using the formula and HCV prevalence of 5%[16]

$$n = \frac{(Z^2\ pq)}{d^2}$$

n = sample size

z = Standard Normal deviation corresponding to 5% level of significance

= 1.96.

p = Prevalence of hepatitis C virus co-infection among patients with chronic liver disease (Chronic Hepatitis B) from review of literature[64]

= 5%. (0.05)

q = complementary probability to p.

1-p

= 1-0.05

= 0.95.

d^2 = precision = 0.05

$$n = \frac{(1.96)^2\ 0.05\ (1-0.05)}{(0.05)^2}$$

The minimum sample size required for this study was 73. However, for the purpose of this study a sample size of 117 was taken to account for loss of questionnaire and loss to follow up.

Sample collection

10 millilitre of venous blood was drawn asepetically into properly labelled sample bottle, allowed to clot, retract and centrifuged at 1500rpm for 15minutes.The sera separated into sample bottle and kept at -20^0C for analysis. Serum transaminases, total protein and bilirubin were analysed using standard routine methods. Serodetection of HbsAg and Anti-HCV was carried out using Biotech diagnostic limited and clinotech diagnostic (ELISA-3), according to the manufacture's instruction. Percutaneous liver biopsy was carried out using the trucut soft tissue biopsy with graduate cannula 14G(2.1mmx152mm) was perfomed on the 100 subjects, Minimum safety requirement of heamoglobin concentration of at least 10g/100ml, platelets count of 80,000/cm^3, prolonged prothrombin time not prolonged than 3 seconds of the control was observed.

Histological analysis

The liver tissues were fixed in 10% buffered normal-saline for 24 hours and embedded in paraffin wax. The liver tissue sections were cut at 5μm thinness with rotary microtome and stained with haematoxylin and eosin. Haematoxylin stains the nuclei while eosin stains the cytoplasm and other extra cellular materials. Special stains such as Reticulin to stain the reticular fibres, Masson's trichrome to demonstrate fibrous connective tissues, a Periodic Acid-Schiff (PAS) to demonstrate glycogen, and Orcein to demonstrate HBsAg were also employed. The histologic activity index of chronic hepatitis were reported according to Knodell and Ishak grading and scoring method.

Data analysis

Using SPSS version 20.0 demographic and clinical data were expressed in descriptive frequency, mean, standard deviation. Categerigical comparison of data was done by X^2 (chi-square) test at significance differences of $p<0.05$. Fisher exact test and multiple logistic regression analysis was performed to assess the independent effects of risk factors for acquisition of HCV. The effects of the risk factors using odds ratio (OR) with 95th CI (confidence interval) were estimated.

Results

The 100 chronic HBV related liver disease study participants (Table 1) comprised 65% males and 35% females giving male to female ratio of 1.9:1, and ratio of 1:1 of study participants aged above >40years. The mean age of study participant was 32.7+9.3years, with majority within the age-group 41-60years(42%). The mean age of co-infected patients was significantly older than monoinfected subjects, 41.2± 8.8 vs 31.8±8.9 years. (P=0.0019).Significant difference was observed between the viral infection and age-group of the study participants(p=0.001)

The seroprevalence of Anti-HCV was 10%, with gender distribution of 10.8%(7/65) males and 8.6%(3/35)females, giving male to females ratio 1:2.3. Of the married patients(48%),18.8%(n=9) had coinfection and 81.2%(n=39) monoinfection, while single participants,98.0%(n=50) had monoinfection and 20%(n=1) coinfection(p=0.118). There was no significant difference between the risk factors and viral infection(p=0.84).

Histological diagnosis of liver biopsies(table 2), chronic hepatitis recorded in 92%cases , 4% in cirrhosis and 4% in hepatocellular carcinoma respectively. There was significant association between Anti-HCV and histological diagnosis with 75% cases identified as HCC, 25% as liver cirrhosis and 6.6% as chronic hepatitis(p=0.01). Using the histological activity index scores graded as <6, 6-10 and >10 of the severity of liver diseases between coinfection and monoinfection, mild necroinflamatory changes of 12.8% vs 87.2%, moderate changes of 12.0% vs 88.0% and severe necroinflammatory 3.6% vs 96.4%. Signification association was observed between the severity of liver diseases and coinfection(p=0.0519), HCC(75% vs 25%) liver cirrhosis(25% vs 75%) and chronic hepatitis(6.6% vs 93.4%) respectively. The mean histological activity index score of coinfected patients was significantly lower than monoinfection 5.2+ 4.8 to 8.0+4.2(p=0.05) Table 3 depicit the biochemical and hematological indices. Biochemical parameters showed significantly high mean serum of AST,ALT, ALP and Total Protein among coinfected patients compared to monoinfected patients(p=0.05),with low serum albumin level. Heamatological indices showed significant difference in the mean PCV, absolute white blood cell count(WBC), platelets count and ESR levels(p=0.05). In coinfected patients, high WBC and ESR was recorded compared to the mean PCV, lymphocyte neutrophil and PLT in monoinfected patients.

Discussion

In this study, the mean age of coinfected patients was higher than monoinfected patients 41.2+8.8years vs 32.12+12.5years,but the mean age(41.2+8.8years) wascomparable to the mean age reported in similar studies, 42.9+12.3years and 47.44+14.56 years in India[17,18], and 42.8+11.5 years in China[19].We observed that majority of coinfected patients were within the age-group of 41-60 years, similar age-group was reported in studied conducted in India, 41-60years [17],41-50years[20] and 30-60years[21]. However, significant association was observed between coinfection and age-group of older patients compared to monoinfection($p<0.0019$), this observation is consistent with some documented studies[19,22]. This association between coinfection and older age-group alluded to the manifestation of liver diseases due to coinfection is common in the fourth decade of life[7,17,19,20].

As observed in most viral hepatitis studies, male gender predominate similar to the picture seen in our study of 65% males compared to 35% females, giving a male to female ratio of 1.9:1[18,20,23].The major driver risk factors of viral factors in SSA are early unprotected sexual activities involving multiple partners, and exposure to risk factors of viral hepatitis infection like transfusion of improperly screened blood units/product, and use of unsterile instrument for invasive procedure like circumcision and uvulectomy [8] . In contrary, we observed no significant difference between the risk factors of viral hepatitis infection and coinfection, which differs from findings ofother similar studies conducted in Indian continent that reported significant association between risk factors- blood

transfusion, alcohol ingestion and intravenous drug ingestion and coinfection[18,24,25].

The coinfection prevalence of 10% is comparable to 11.0 % and 12.8% reported instudies conducted in Spain[26,27], but lower than 18% in China[19], 16% in India[28], 14.4% in Maiduguri Northeast Nigeria[10], and 12.2% in Southwest Nigeria[29]. While other studies had reported lower prevalence, 4% in India[17], 5.9% in India [30] and 7.7% in Mongolia [31].This observed variation may be due to several factors, such as method employed in the study, criteria of inclusion of patients selection and chronicity of liver diseases.

The clinical usefulness of histological diagnosis of liver biopsies in investigation and assessment of liver diseases has been well documented[10,11,15,29,32]. However, the unique characteristic is the difference in the histological picture and severity/CLD subtypes [15]. In our study, histological finding of liver biopsies revealed 92% cases were identified as chronic hepatitis and 4% each as liver cirrhosis and HCC cases respectively. Further evaluation of coinfected liver biopsies, revealed that 60% cases were confirmed as chronic hepatitis, 30% liver cirrhosis and 10% HCC respectively. Comparing our histological findings with other similar studies revealed variation in the chronic liver disease subtypes as reported in other studies. In northeastern Nigeria, Laraba*et al* reported, 20.5% prevalence of HCC, 12.1% cirrhosis and 5.6% chronic hepatitis. While in India, Hossani*et al*reported cirrhosis(30%), HCC(26%) and chronic hepatitis(8%)[20], cirrhosis(20%), and 30% chronic hepatitis[33]. In China, Dai *et al* reported 67% cases of chronic active hepatitis followed by 22% chronic persistent and 11% liver cirrhosis. A multicenter study of 9,997 cases in Italy, Sagnelli*et al*[15] reported 62.1% chronic hepatitis, 19.4% liver cirrhosis and 3.4% HCC.

The Anti-HCV prevalence of coinfectedbiopies revealed 75% in HCC, 25% in liver cirrhosis and 6.6% in chronic hepatitis. Studies with similar high Anti-HCV prevalence includes, 75% in Spain[34], 69.6% in Spain[35] and 65 % in Italy[36]. Some studies has also reported low Anti-HCV prevalence in HCC cases , 18% in China[19], 20% in Maiduguri[10], 12.2% in Lagos[11], 14% in Southwest Nigeria[37], 18.7% in Ibadan, and 19% in The Gambia[32]. The non-significant difference observed in histological diagnosis between coinfected(75% in HCC) and monoinfected (chronic hepatitis 93.4%, cirrhosis 75.0%) in liver biopsies, alluded to the fact that coinfection may not necessarily aggravate liver damage[38]. Though, some studies have associated high prevalence of cirrhosis, chronic liver disease with HCC cases[39,40] The reason for the histological picture recorded in our study may be due to HBV endemicity that could be responsible for possible superinfection, and criteria of patient selection.

Several studies have reported high prevalence of CLD subtypes, liver cirrhosis and chronic liver disease in coinfection than monoinfection[41,42], with high prevalence of Anti-HCV and severity of chronic HBV infection[43]. However, severity of liver disease in coinfection and monoinfection is debatable . In our study, we observed significant decrease in the mean HAI score of coinfections(52.+4.8) compared to monoinfection(8.2+4.2), comparable to other studies[15,44], this observed activity may be due to severity of liver disease which is a reflection of progression from mild to severe necroinflammatory chronic hepatitis, to cirrhosis and HCC. Using the Knodellclassification, we observed mild presentation of 12.8% in coinfection compared to mono infection of 87.2%, moderate 12.0% vs 88.0% and severe case 3.6% vs 96.4%. However, the highnecroinflammmatory changes observed in monoinfection cases may be due to

relatively low HCV prevalence in the study area, or possible HBV superinfection, capable of development and progression from cirrhotic to fibrosis stages.

Biochemical parameters are used as clinically predictors of assessing viral hepatitis disease and antiviral therapy, and thevalues varies with severity of liver disease. However, ALT is one of the important biochemical tests in assessment of CLD but is a poor marker when considering antiviral therapy, as it shows poor correlation in patients with significant liver injury in chronic HBV infection. In this study, significantly high mean values of ALT, AST, ALP, total Protein and reduced albumin level was recorded, which is in conformity with the findings of other studies[15,19, 20,21,45-47]. The increased liver enzyme activities recorded in coinfection is associated with liver disorder, which is associated with decreased serum albumin[47,49].

Heamtological parameters assessed the heamopoetic system response to viral infection[50]. In this study, significantly high mean value of WBC and ESR was recorded in coinfected patients, in contrast to significant mean PCV, lymphocyte and PLT counts in monoinfection. The mean hematocrit value of 39.0 vs 39.26 in coinfection and monoinfection, showed that 13%(<33%) coinfected patients were anaemic, which is lower than 24% reported in acute viral hepatitis infection[50]. In this study, we observed increased mean absolute WBC(7983X 10^9), and 5% leucocytosis in coinfection, these values are lower than 12% reported in acute hepatitis study in Southwest Nigeria[50].The mean lymphocyte count of 44.18% in monoinfection is a indication of viral infection[50,51]. Thrombocytopenia is a common heamatological picture in chronic liver disease, which the count could vary with the severity of infection, from mild to severe cases[52]. In our study, increased mean PLT (403,516) was observed in monoinfection compared to coinfection of (394,757), this observed PLT count in both coinfection and

monoinfection may be due to chronic infection, malignancy, chronic inflammatory disease or iron deficiency [52] The significant difference observed in the mean PCV and ESR, is consistent to the report of Ajugwo and Ukaji [53] study which reported low PCV and ESR, indicative of inflammatory liver disease. The main findings of our study revealed concomitant contribution of both aetiological viruses in development and progression of chronic liver diseases and changes in laboratory parameters necessary for patient assessment and management approach

Despite the fact that the study findings had presented clinical, pathological and epidemiological information, necessary for better understanding and viral hepatitis infection in a low-resource setting.. There are limitations, (i) the non-serodetection of HBeAg, an indicator of infectivity and (ii) molecular assay of HBV-DNA and HCV-RNA not included in the study had limited the vital immunological information necessary in evaluating the stage of infection and possible seroconversion.

Conclusion

The coinfection prevalence of 10% is relatively lower than reported in other studies, which depict disparity and variation due to factors influencing infection outcome. The chronic liver disease subtypes prevalence was similar to those reported in other studies. However, the histological diagnosis of liver biopsies showed no significant difference in degree of severity which highlight that coinfection might not necessarily impact on liver damage.

References

1-Marianna G. Mavilia and George Y. WuHBV-HCV Coinfection: Viral Interactions, Management, and Viral ReactivationJournal of Clinical and Translational Hepatology 2018 vol. 6 | 296–305

2-World Health Organization. New hepatitis data highlights need for urgent global response. Geneva (CH): World Health Organization; 2017. Available at: http://www.who.int/mediacentre/news/releases/2017/global-hepatitis-report/en/.

3-Perz JF, Armstrong GL, Farrington LA, Hutin YJ, Bell BP. The contributions of hepatitis B virus and hepatitis C virus infections to cirrhosis and primary liver cancer worldwide. *JHepatol*2006; **45**: 529-538 [PMID: 16879891]

4- Raimondo G, Brunetto MR, Pontisso P, Smedile A, MainaAM,Saitta C, et al. Longitudinal evaluation reveals a complex spec-trum of virological profiles in hepatitis B virus/hepatitis Cvirus-coinfected patients. Hepatology 2006;43(1):100—7.

5-Pol S, Haour G, Fontaine H, Dorival C, Petrov-Sanchez V, Bourliere M, et al. The negative impact of HBV/HCV coinfection on cirrhosis and its consequences. Aliment PharmacolTher2017;46:1054–1060. doi: 10.1111/apt.14352.

6- Chu CJ, Lee SD. Hepatitis B virus/hepatitis C virus coinfection: Epidemiology, clinical features, viral interactions and treatment. J GastroenterolHepatol2008;23:512−20.

7- Tyson GL, Kramer JR, Duan Z, Davila JA, Richardson PA, El- Serag HB. Prevalence and predictors of hepatitis B virus coinfection in a United States cohort of hepatitis C virus infected patients. *Hepatology*2013;**58**:538-545

8-Olayinka AB, Oyemakinde B, Balogun MS, Ajudua A, Nguku P, Aderinola M, Egwuenu-Oladejo A, et al Seroprevalence of Hepatitis B Infection in Nigeria: A National SurveyAm. J. Trop. Med. Hyg., 95(4), 2016, pp. 902–907

9-BM Musa, S Bussell , MM Borodo, AA Samaila, OL Femi Prevalence of hepatitis B virus infection in Nigeria, 2000-2013: A systematic review and meta-analysis Nigerian Journal of Clinical Practice • Mar-Apr 2015 • Vol 18

10-Laraba A, Wadzali G, Sunday B, et al., Hepatitis C Virus Infection in Nigerians With Chronic Liver Disease. Internet J Gastroenterol 2009; 9(1):1-3.

11-Lesi OA, Kehinde MO, Anomneze EE. Chronic liver disease in Lagos: a clinicopathological study. Niger Postgrad Med J 2004; 11: 91-6

12-Shehu MY. Prevalence of hepatitis C virus antibodies among patients with chronic liver disease at the Jos University Teaching Hospital. A dissertation submitted to the West African College of Physicians in partial fulfillment of the requirements for the award of Fellowship of the bb college, 2002

13-Villari D, Pernice M, Spinella S, et al. Chronic hepatitis in patients with active hepatitis B virus and hepatitis C virus combined infections: a histological study. *Am J Gastroenterol*1995;**90**:955-958.

14-Colombari R, Dhillon AP, Piazzola E *et al.*Chronic hepatitis in multiple virus infection: histopathological evaluation. *Histopathology* 1993; **22**: 319–25.

15- E. Sagnelli, G. Pasquale, N. Coppola, F. Scarano, C. Marrocco, C. Scolastico,T.Santantonio, A. Gentile, F. PiccininoInfluence of Chronic Coinfection with Hepatitis B and C Virus on Liver HistologyInfection 2004; 32: 144–148

16-Wesly A. Altar MJ: Epidemiology of hepatitis C: Geographical differences and temporal trends. Semin liver dis 2000; **20**:1.

17- Grewal US, Walia G, Bakshi R, Chopra S. Hepatitis B and C Viruses, their coinfection and correlations in chronic liver disease patients: A tertiary care hospital study. Int J App Basic Med Res 2018;8:204-9.

18Amarjit Kaur Gill, Vivek Mittal, Vanita Mittal, Narinder Kaur, Amandeep Kaur, A K Maria, Rajinder Singh Seroprevalence of HBsAg, anti-HCV and co-infection among liver disease patients: a prospective observational trial. International Journal of Contemporary Medical Research 2016;3(8):2204-2208.

19-Dai CY, Yu ML, Chuang WL *et al.* Influence of hepatitis C virus on the profiles of patients with chronic hepatitis B virus infection. *J. Gastroenterol. Hepatol.* 2001; 16: 636–40

20-Hossani MM, Khandaker MAK, Azad KAK, SardarMH , Deb SR, Mallik MU. Clinicopathological correlation of liver biopsy- study of 50cases *J Dhaka Med Coll. 2013; 22(2) : 120-124.*

21-Prabha Desikan1, Aseem Rangnekar1, Zeba Khan1, Nikita Panwalkar1, Protiti Bose1, HanniVasudev Gulwani2, Sukhpreet Kaur2. Sero-Occurrence of HBV/HCV Co-infection and Levels of Liver Enzymes among Patients at a Tertiary Care Hospital in Central India: a Pilot Study. Central Asian Journal of Global HealthVolume 8, No. 1 (2019)

22-Ohkawa K, Hayashi N, Yuki N, Hagiwara H, Kato M, Yamamoto K, Eguchi H, Fusamoto H, Masuzawa M, KamadaT.Hepatitis C virus antibody and hepatitis C virus replication in chronic hepatitis B patients. *J Hepatol*1994; **21**: 509-514

23- Chowdhury AW. A Study of HBsAg status of hospital admitted patients of chronic liver disease and its clinicopathological correlation [Dissertation]. Dhaka: Bangladesh College of Physicians and Surgeons (BCPS); 1995

24- Singh V, Katyal R, Kochhar RK, Bhasin DK, Aggarwal RP. Study of hepatitis B and C viral markers in patients of chronic liver disease. Indian J Med Microbiol2004;22:269−70.

25- Devi KS, Singh NB, Mara J, Singh TB, Singh YM.Seroprevalence of hepatitis B virus and hepatitis C virus among hepatic disorders and injecting drug users in manipur – A preliminary report. Indian J Med Microbiol2004;22:136−7.

26- Fong T.L, Di Bisceglie AM, Waggoner JG, *et al*. The significance of antibody to hepatitis C virus in patients with chronic hepatitis B. Hepatology 1992; **14**:64 -7.

27- Crespo J, Lozano J C, de la Cruz F *et al*. Prevalence and significance of hepatitis C. Viremia in chronic active hepatitis B. AM. J. Gestroenterol 1994;**89**:1147-51

28-Chakravarti A, Verma V, Jain M, Kar P. Characteristics of dual infection of hepatitis B and C viruses among patients with chronic liver disease: a study from tertiary care hospital. Trop Gastroenterol 2005;26(4):183–7.

29-Olubuyide IO, Aliyu B, Olaleye OA, Ola SO, Olawuyi F, Malabu UH et al. Hepatitis B and C virus and Hepatocellular carcinoma. Trans R Soc Trop Med Hyg 1997;91(1): 38-41

30-Saravanan S, Velu V, Nandakumar S, Madhavan V, Shanmugasundaram U, Murugavel KG, Balakrishnan P, Kumarasamy N, Solomon S, Thyagarajan SP. Hepatitis B virus and hepatitis C virus dual infection among patients with chronic liver disease. *J MicrobiolImmunol Infect* 2009; **42**: 122-128

31-Tsatsralt-Od B, Takahashi M, Nishizawa T, Endo K, Inoue J, Okamoto H. High prevalence of dual or triple infection of hepatitis B, C, and delta viruses among patients with chronic liver disease in Mongolia. *J Med Virol*2005; **77**: 491-499

32- Kirk GD, Lesi OA, Mendy M *et al.* The Gambia Liver Cancer Study: Infection with hepatitis B and C and the risk of hepatocellular carcinoma in West Africa. *Hepatology*2004; 39: 211–19.

33-Anderson E, Ridly DS. Liver biopsy and precutaneous cholangiography using posterior approach 500 needle biopsy. Am J Surg 2003; 121: 31-42.

34-Bruix J. Barnera Jm, Calvet x *et al*. prevalence of antibodies to hepatitis C virus in Spanish pa;tients with hepatocellular carcinoma. Lancet 1989; **2:** 1004-1009.

35-Yuki N, Hayeshi N, Kasahare A *et al* hepatitis B virus maker and antibodies to hepatitis C virus in Japan patients with hepstocellular carcinoma. Digestive Diseases and Science 1992; **37:** 65-72.

36-Colombo M, Kuo G; Choo QL *et al*. Prevalence of anti bodies to hepatitis C virus in Halian patients with hepstocellular carcinoma. Lancet 1989; **2:**1006-1008.

37-. Ola SO, Odaibo GN, Olaleye OD. HCV and HBV infections in Nigerian patients with Liver cirrhosis and Hepatocellular carcinoma. Nig Q J Hosp Med 2004;14( ):

38-Konstantinou D, Deutsch M. The spectrum of HBV/HCV coinfection: epidemiology, clinical characteristics, viralinteractions and management. Ann Gastroenterol2015;28:221–228.

39- Huang YT, Jen CL, Yang HI, Lee MH, Su J, Lu SN, Iloeje UH, Chen CJ. Lifetime risk and sex difference of hepatocellular carcinoma among patients with chronic hepatitis B and C. *J ClinOncol*2011; **29**: 3643-3650 [PMID: 21859997 DOI:10.1200/JCO.2011.36.2335]

40-Aghemo A, Colombo M. Treatment of patients with dual hepatitis B and C: a step in the right direction. *Gut* 2014; 63:

41- Lee LP, Dai CY, Chuang WL *et al.* Comparison of liver histopathology between chronic hepatitis C patients and chronic hepatitis B and C-co-infected patients. *J. Gastroenterol. Hepatol.* 2007; **22**: 515–17.

42- Z. Liu and J. Hou, "Hepatitis B virus (HBV) and hepatitis C virus (HCV) dual infection," i*nternational Journal of Medical Sciences*,vol. 3, no. 2, pp. 57–62, 2006.

43- C. H. Chen, P. M. Yang, G. T. Huang, H. S. Lee, J. L. Sung, and J. C. Sheu, "Estimation of seroprevalence of hepatitis B virus and hepatitis C virus in Taiwan from a large-scale survey offree hepatitis screening participants," *Journal of the Formosan Medical Association*, vol. 106, no. 2, pp. 148–155, 2007

44-Marrot A, Belaid A, Orlent H, Sersté T, Michielsen P, Colle I, et al. Characteristics of patients with hepatitis B virus and hepatitis C virus dual infection in a Western European country: Comparison with monoinfected patients. Clin Res HepatolGastroenterol2017;41:656–663.

45-Zarski JP, Bohn B, Bastie A, Pawlotsky JM, Baud M, Bost-Bezeaux F, et al.Characteristics of patients with dual infection by hepatitis B and C viruses.JHepatol 1998;28:27–33.

46-Senturk H, Tahan V, Canbakan B, Dane F, Ulger Y, Ozaras R, et al. Clinicopathologic features of dual chronic hepatitis B and C infection: a

comparison with single hepatitis B, C and delta infections. Ann Hepatol 2008;7(1):52–8

47-Abdullah SM. Prevalence of Hepatitis B and C virus infection and their co-relation with hematological and hepatic parameters in subjects undergoing Premarital Screening in the Jazan Region, Kingdom of Saudi Arabia. Pak J Med Sci. 2018;34(2):316-321.

48- PrabhaDesikan, AseemRangnekar, Zeba Khan, Nikita Panwalkar, Protiti Bose, HanniVasudevGulwani, Sukhpreet Kaur. Sero-Occurrence of HBV/HCV Co-infection and Levels of Liver Enzymes among Patients at a Tertiary Care Hospital in Central India: a Pilot Study. Central Asian Journal of Global Health Vol. 8, No. 1 (2019)

49-Garcovich M, Zocco MA, Gasbarrini A. Clinical use of albumin in hepatology. Blood Transfus. 2009;7(4):268-277.

50- Fasola FA, Otegbayo JA, Abjah UMA and Ola SA. Heamatological parameters in Nigerians with acute viral hepatitis. *Nigerian Journal of Gastroenterology and Hepatology Vol.1 No1 June, 2009* 27-31

51— Mohamed AE, Ven D, Al Karawi MA. Dual infection with hepatitis C and B viruses: clinical and histologic study in Saudi patients. Hepatogastroenterology 1997;44:1404–6.

52-Nezam Afdhal, John McHutchison2 , Robert Brown3 , Ira Jacobson4 , Michael Manns5 , Fred Poordad6 , Babette Weksler4 , Rafael Esteban7 1

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53-Ajugwo, A. O. and Ukaji, D. C. (2015).SomeHaematological Parameters of Symptomatic and Asymptomatic Hepatitis B Positive Patients Attending a Nigerian Tertiary Hospital. British Journal of Medicine and Medical Research. 7 (3): 219-223.

Table 1; Demographic variable of study patients

Variables	Coinfection	Monoinfection	p-value
Mean age(SD)(yrs)	42.0+8.8	31.8+8.9	
Age-group			
<20	1(14.3)	7(85.7)	
21-30		42(100)	
31-40	2(6.9)	27(93.1)	0.0019
41-50	6(37.5)	10(62.5)	
>51	1(20.0)	4(80.0)	
Sex			
Male	7(10.8)	58(89.2)	0.001
Female	3(8.6)	32(91.4)	
Marital status			
Divorce/widowed	0(0)	1(100)	
Married	9(18.8)	39(81.25)	0.116
Single	1(1.95)	50(98.03)	

Risk factors			
Blood transfusion	1(9.1)	10(90.9)	
Multiple sex partners	2(15.4)	11(84.6)	0.84
Traditional incision	1(6.3)	15(93.7)	
Uvulectomy	2(14.3)	12(15.7)	
Histopathological diagnosis			
Chronic hepatitis	6(6.6)	86(93.4)	
Liver cirrhosis	1(25.0)	3(75.0)	0.01
Hepatocellular carcinoma	3(75.0)	1(25.0)	

Table 2; Histological diagnosis and Knodell scores of Liver biopsies

Chronic Liver disease	HCV(+)	HCV (-)	P=value
Chronic hepatitis	6(6.6%)	86(93.4)	
Liver Cirrhosis	1(25.0%)	3(75.0)	0.01
HCC	3(75.0%)	1(25.0%)	
Knodell Scores			
<6	6(12.8%)	41(87.2)	
6-10	3(12.0)	22(88.0)	0.0519
>10	1(3.6)	27(96.4)	

Table 3 **Biochemical and Heamatological parameters of dual and monoinfected patients**

Parameters	HBV +HCV	HCV(-)	p-value
ALP(IU/L)	110.9	72.0	0.045
ALT(IU/L)	26.7	9.8	0.015
AST(IU/L)	43.3	12.0	0.045
Total Protein(g/L)	81.7	72.3	0.012
Total Bilirubin(umol/L)	13.0	13.6	0.84
Albumin(g/L)	40.8	39.1	0.52
PCV	39.26	39.00	0.047
WBC	7983	7212	0.013
Neutrophil	50.38	49.90	0.08
Lymphocyte	43.87	44.18	0.012
PLT	394,757	403,516	0.001
ESR	10.96	11.03	0.011