AFRICAN JOURNAL OF MEDICINE and medical sciences

VOLUME 35 NUMBER 3 SEPTEMBER 2006

ISSN 1116—4077

Afr J Med Med Sci (2006) 35, 337-343

Seroprevalence and biochemical features of hepatitis B surface antigenemia in patients with HIV-1 infection in Lagos, Nigeria

BA Iwalokun[1]; SO Hodonu[2]; BM Olaleye[1] and OA Olabisi[1].

[1]*Department of Biochemistry, Lagos State University, PMB 1087, Apapa – Lagos.* [2]*Faculty of Clinical Sciences, Lagos State University College of Medicine, PMB. 21266, Ikeja –Lagos, Nigeria*

Summary

Studies have confirmed HBsAg antigenemia as an important correlate of HIV-1 infections in Nigeria. However, the hepatic pathophysiology and therapeutic implications in antiretroviral regimens are poorly understood. In this study, fifty-four HIV-1 seropositive patients aged 16 – 47 years (mean age 31.8 years) with CD4 T lymphocyte counts of 148 – 420 cells/mm^3 attending clinics in General Hospital, Ikeja and private medical centres in Lagos Island, Nigeria and forty sex and age-matched apparently healthy controls were serologically examined as carriers of hepatitis B surface antigen (HBsAg) using a particle agglutination assay procedure (Sensitivity 94.5 – 100%, Serodia®-HBs.PA, Fujirebio. Inc.). HBsAg was detected in 28 (51.9%) and 5 (12.5%) of the patients and controls respectively ($\pm$ 2 Mantel-Haenszel = 13.8; P = 0.02). HBV co-infection was found to result in significant (P < 0.05) reduction in total lymphocyte count (1368.6 $\pm$ 53.2 vs. 1590.5 $\pm$ 80.4 cells/mm^3) with 7 of 10 (70%), 18 of 33 (54.5%) and 3 of 6 (50%) HIV-1 patients having < 200, 200 – 350 and > 350 CD4 lymphocyte cells/μl and eliciting HBsAg antigenemia. These patients exhibited 2.9 – 8.6% reduction in CD4 T lymphocyte counts compared to their seronegative counterparts. Although the liver function parameters measured in HIV-1 patients tested were higher than control values, significantly (P < 0.05) elevated liver enzymes: sGOT (44.1 $\pm$ 2.2 vs. 26.2 $\pm$ 2.1 IU/L), sGPT (46.2 $\pm$ 2.4 vs. 23.5 + 1.8 IU/L), and serum bilirubin levels (2.04 $\pm$ 0.18 vs. 1.0 $\pm$ 0.07 mg/dL) were observed in HBsAg positive HIV-1 patients. sGOT or sGPT activity that was five times greater than the control was observed in 7(25%) and 2 (7.7%) of HbsAg positive and negative HIV-1 patients in whom significant association between decreased total lymphocyte count and measured liver parameters was found. We conclude that hepatitis infection deteriorates liver functions and its investigation in HIV-1 infected patients may be of clino-therapeutic importance prior to antiretroviral therapy administration.

Keywords: *Hepatitis B surface antigen (HBsAg), HIV – 1 infection, CD4 T lymphocyte count, therapeutic implications.*

Correspondence Dr B A Iwalokun, Department of Biochemistry, Lagos State University PMB 1087, Apapa, Lagos, Nigeria. Email bamwal@yahoo.com

Résumé

Les études ont confirmées l'antigénique HBsAg comme un important indicateur des infections du HIV au Nigéria. Cependant la pathophysiologie hépatique et les implications des régiment antiretroviraux sont de moins compris. Dans cette étude, 54 patients séropositif agé de 16 – 47 ans (moyenne d'age 31.8 ans) avec des taux de lymphocytes CD4 de 148-420 cellules /mm2 participaient a la clinique de l'hôpital général d'Ikeja et les centres prives de Lagos et 47 individus comme contrôle sain étaient examinés serologiquement comme porteur d'hépatique B. 28(51.9%) avaient HBsAg et 5(12.5%) des patients et contrôle respectivement ; La co- infection du HBV influençait significatiment la réduction totale des taux des lymphocytes sur 7 sur 10(70%) (136.6 $\pm$ 53.2 vs 1598.5 $\pm$ 80.4 cellules/mm 3),18 sur 33 (54.5%) et 3 sur 6 (50%) ayan le VIH-1 ayant des taux de CD4 de <100,200-350, >350 cellules/μl avec l'HBsAg antigénique. Ces patients démontraient un taux de réduction de 2.9-8.6% comparé aux contrôles sains. Bien que les paramètres des fonctions du foi mesurés chez ces patients ayant le VIH-1 avaient des valeurs cinq fois plus élevé qu'au contrôle sain en activité des enzymes hépatiques (Sgot (25%) vs Sgpt (7.7%)) et taux de sérum bilirubine.Ily a avait une corrélation significative entre les taux de lymphocytes et les paramètres de fonctions hépatiques. Nous avons conclu que l'hépatite détérioré les fonctions du foi et les investigations sur les patients ayant le VIH-1peuvent être d'importance clino-therapeutique avant l'administration des médicaments antiretroviraux.

Introduction

Infection with hepatitis virus (HBV) alone accounts for approximately 350 million chronic carrier cases every year worldwide [1]. In the last two decades, deadly combination of HBV and human immunodeficiency virus (HIV) infection has emerged and increased significantly to accelerate the progression of infections to clinical Acquired Immune Deficiency Syndrome (AIDS), liver cirrhosis and hepatocellular carcinoma with morbidity and mortality eventualities in children, pregnant women and health workers [2, 3, 4].

In Nigeria, HBV infections due to genotype E strains with escape mutations have been reported [5] and several studies have demonstrated high risk of HBV acquisition in laboratory and hospital workers due to inad-

equate precautionary facilities and awareness of risk factors of transmission [6,7]. The prevalence of hepatitis B surface antigen (HBsAg), which signifies HBV infection [8] has also been observed among patients with liver cirrhosis and hepatocellular carcinoma in many Nigerian hospitals [9]. A survey of cancer-associated deaths among Nigerian children during the period 1965 – 1985 reported HBV as the major aetiological agent [10]. The shared epidemiological risk factors by HBV and HIV have also enabled co-infection possibilities with only few of such cases reported in Nigeria [11,12].

In addition to limited access to antiviral therapies in the country, regimens are also constrained by narrow alternatives eliminating the possibility of protease inhibitor (PI) switch within regimens based on efficacy and tolerance. This has been the practice in some countries with HIV/HBV endemia [13]. PIs are hepatotoxic and the hepatic cytolysis that they induce has been found to be accelerated by HBV/HIV co-infection [14]. Thibault *et al* [15] also reported rapidity in the acquisition of lamivudine resistance induced by several point mutations in the genomes of HIV and HBV variants in sick patients. While in the work of Davis [16], poor clinical response to interferon alpha in patients with HBV infection was attributed to HIV-1 viremia.

While the impact of the integration of hepatitis B vaccination in extended programme on immunization as a control measure against HBV infection and its sequelae has been successful in some countries [17,18], the unclear understanding of the pathophysiology of HBV/HIV co-infection coupled with paucity of data on the clinical implications arising from this deadly combination have allowed doubts on the clinical efficacy and effectiveness of antiviral regimens to stay and questions on when to administer HAART or effect PI switch to still be a clinically important decision a physician has to make. Data on hepatic cytolysis status of HIV –infected patients, primarily characterized by alanine transaminase activity five times greater than normal level on which PI – switch or withdrawal is based [19] is lacking in Nigeria.

The present study was designed to determine the prevalence of HBsAg seropositivity among HIV-infected patients in Lagos, Nigeria. Effects of co-infection on liver function parameters, total lymphocyte and CD4 T lymphocyte counts were also evaluated.

Materials and methods

Subjects

A total of fifty-four (54) inpatients and outpatients (Male/Female = 30/24) aged 16 – 47 years (mean age = 31.8 years), newly diagnosed for Human Immunodeficiency virus type 1 (HIV-1) infection and attending private hospitals and medical clinics of General Hospitals Ikeja and Lagos were enrolled into the study. Data such as age, sex, and year of diagnosis were obtained from each patient with the aid of a questionnaire. HIV-1 detection was based on immunoassay detection of HIV-1 seropositivity and confirmation by western blotting carried out in the Biomedical Laboratory of the Nigerian Institute of Medical Research (NIMR) and Central Public Health Laboratories. Yaba, Lagos. Data on baseline of CD4 T lymphocyte counts of the patients were extracted from their case report form.

Forty (40) apparently healthy individuals comprising 20 males and 20 females and aged 16 – 42 years (mean age = 28.6 years) were also recruited into the study as controls. Informed consent was obtained from each subject in the cohort. Ethical clearance was obtained from the Hospital Management Board, Lagos, Nigeria.

All the study participants were serologically screened for the presence of hepatitis B virus surface antigen (HBsAg) in their sera. The latter were obtained by centrifuging whole blood collected in plain bottles at 1,500 x g for 5 min. Serum liver enzyme activities and bilirubin level were also determined. Aliquots of blood samples in EDTA bottle were used for total leukocyte and lymphocytes enumeration.

Detection of serum hepatitis B surface antigen

HBsAg, a biomarker of acute or chronic hepatitis B infection was detected in serum by a qualitative particle agglutination assay [Serodia®-HBs.PA (Fujirebio.Inc)] in a 96-well microtitre plate. The sensitivity of true positive results indicated by agglutination after 30 - 60 seconds of incubation ranged from 94.5 – 100% [20].

Haematology

Total leukocytes in blood were determined microscopically in Neubauer counting chamber according to Dacie and Lewis [21]. Total lymphocyte count was calculated by multiplying percentage lymphocyte count on Leishman's stained blood films by total leukocyte count.

Biochemistry

Activities of serum glutamatic-pyruvic transaminase (sGPT) and serum Glutamate – oxaloacetate transaminase (sGOT) were determined spectrophotometrically according to Bergmeyer and Bent [22]. Serum alkaline phosphatase (ALK) activity was measured as the rate of release of inorganic phosphate (Pi) from p-nitrophenol phosphate (PNP) according to Denier *et al* [23]. Serum lactate dehydrogenase activity based on $NADH - NAD^+$ interconversion was estimated by using the protocol adopted by Wroblewski and Ladue [24]. Serum total bilirubin level was determined spectrophotometrically as described by Jendrassik and Grof [25].

Statistical analyses

Data were analyzed as mean $\pm$ standard error of mean (SEM) and percentages. Disparity between the mean values of measured parameters in HIV-1 infected and HIV/HBV co-infected patients and the controls were analyzed

by one-way analysis of variance (ANOVA). Differences observed in percentages were evaluated using chi-square analysis with Mante-Haenszel modification in the STATCALC program of EPI-INFO version 6 statistical software (CDC, Atlanta, GA). Multivariate regression analysis was used to evaluate the magnitude of associations between parameters. P values less than 0.05 were considered significant and P values greater than 0.05 were not significant.

Results

Carriage of hepatitis B surface antigen (HBsAg) among HIV-1 infected patients and apparently healthy control subjects.

In this study, a total of fifty-four (54) HIV-1 seropositive patients and forty (40) apparently healthy controls were screened for the presence of serum HBsAg, a marker of HBV infection. The prevalence rates of HBsAg antigenemia were found to be 51.9% (28 of 54) and 12.5% (5 of 40) respectively in HIV-1 infected patients and the controls (χ^2Mantel-Haenszel = 13.8; P = 0.02) (Table 1).

Table 1. Carriage of hepatitis B surface antigen (HBsAg) among HIV-1 infected patients and apparently healthy controls.

Subjects	n	HBsAg seropositivity (+)	(-)
HIV-1	54	28	26
Control	40	5	35

*χ2Mantel-Haenszel = 13.8; P = 0.02**
n = Sample size, n (%) = χ^2 = chi-square analysis with Mantel-Haenszel modification.
**Significant difference in prevalence rates of HBsAg antigenemia (HIV-1 versus Control).*

Blood lymphocytes levels and distribution in HIV-1 infected patients and the controls.

Although the difference in their mean ages (32.3 ± 1.7 vs. 29.2 ± 1.3 years) was not significant (P = 0.17), the total lymphocyte count in co-infected patients (1368.6±53.2 cells/mm^3) was found to be significantly ($P < 0.05$) lower than 1590.5± 80.4 cells/mm^3 in HBsAg seronegative HIV-1 patients. Both mean values were in turn significantly ($P < 0.05$) lower than 1820.4 ± 34.7 cells/mm^3 observed in the controls. CD4 T lymphocyte profiling of the HIV-1 infected from the 49 of the 54 case forms analyzed showed that 7 of 10 (70%), 18 of 33 (54.5%) and 3 of 6 (50%) patients with < 200 (158.3 ± 7.4 vs. 172.7 ± 6.8 cells/μL; P = 0.05), 200 – 350 (279.1 ± 9.5 vs. 287.5 ± 8.9 cells/μL; P = 0.07) and > 350 cells /μL (378.1 ± 13.3 vs. 413.5 ± 5.5 cells /μL; $P = 0.05$) were HBsAg seropositive and had 2.9 – 8.6% reduction in counts compared to their seronegative counterparts (Table 2).

Serum levels of liver function parameters among the HBsAg seropositive and seronegative HIV-1 infected patients.

The data presented in Table 3 revealed significant ($P < 0.05$) increases in the serum activity of sGOT, sGPT, LDH, and ALK as well as bilirubin levels in HIV-1 infected and co-infected patients when compared to the control ($P < 0.05$). However, levels of SGOT in HBV/HIV co-infected patients (44.1 ± 2.2 IU/L) were found to be 68.3% higher than 26.2 ± 2.1 IU/L ($P = 0.01$) in HIV-1 patients. The HBsAg seropositive group also exhibited significant ($P < 0.05$) 50.5 – 96.67% increases in serum activity of SGPT (46.2 ± 2.4 vs. 23.5 ± 1.8 IU/L), LDH (293.8 ± 4.6 vs. 191.4 ± 5.6 IU/L) and alkaline phosphatase (201.3 ± 4.2 vs. 133.7 ± 6.1 IU/L). Serum levels of total biliribun ranged from 0.9 – 4.3mg/dL and 0.7 – 1.6mgdL in co-infected and HBsAg seronegative patients respectively with the former exhibiting 104.1% icteric elevation (2.04 ± 0.18 vs. 1.0 ± 0.07mg/dL; $P = 0.01$). 12 (42.9%) of 28 HBsAg seropositive patients and 2 (7.7%)

Table 2. Blood lymphocyte levels and distributions in HIV-1 infected patients and the controls.

Parameters	HIV-1 patients HBsAg (+ve)	HBsAg (-ve)	Control	P
Age (years)	32.3 ± 1.7	29.2 ± 1.3	28.6 ± 2.3[b]	0.17
‡Blood lymphocyte	1368.6 ± 53.2	1590.5 ± 80.4	1820.4 ± 34.2[a]	0.03
‡CD4 T Lymphocytes (n = 49)				
< 200 (n = 10)	158.3 ± 7.4 (n = 7)[83]	172.7 ± 6.8 (n = 3)	ND	0.05
200 – 350 (n = 33)	279.1 ± 9.5 (n = 18)[29]	287.5 ± 8.9 (n = 15)	ND	0.07
> 350 (n = 6)	378.1 ± 13.3 (n = 3)[86]	413.5 ± 5.5 (n = 3)	ND	0.05

Figures in parentheses represent number (n) of cases. Data are given as mean ± SEM and analyzed by one-way ANOVA using F-statistics.
‡Lymphocyte counts are measured in cells/mm^3 with figures in superscript representing % reduction in counts calculated based on HBsAg (-ve) values set at 100%. P = exact probability values for (HBsAg (+ve) vs. HBsAg (-ve). [a]P < 0.05 (HIV-1 vs. Controls). [b]P > 0.05 (HIV-1 vs. Controls). ND = not determined.

Table 3: Serum levels of liver function parameters among the HBsAg seropositive and seronegative HIV-1 infected patients.

Parameters	HIV-1 patients HBsAg (+ve)	HBsAg (-ve)	Control	P
sGOT (IU/L)	44.1 ± 2.2 [68.3]	26.2 ± 2.1	10.6 ± 1.1[a]	0.01
sGPT (IU/L)	46.2 ± 2.4 [96.6]	23.5 ± 1.8	10.1 ± 1.4[a]	0.01
LDH (IU/L)	293.8 ± 4.6 [53.5]	191.4 ± 5.6	184.5 ± 3.7[a]	0.01
ALK (IU/L)	201.3 ± 8.3 [50.6]	133.7 ± 6.1	98.4 ± 5.1[a]	0.01
TB (mg/dL)	2.04 ± 0.18 [106.1]	1.0 ± 0.07	0.82 ± 0.07[a]	0.01
@TB > 2.1mg/dL n(%)	12(49.2)	2(7.7)	0(0)	0.05

Data given in mean ± SEM were analyzed by one- way ANOVA using the F- statistics and difference in proportions was evaluated by $\chi 2$ analysis with Mantel-Haenszel modification. Figures in superscript representing % increase in values calculated based on HBsAg (-ve) values set at 100%. P = exact probability values for (HBsAg (+ve) vs. HBsAg (-ve). [a]P < 0.05 (HIV-1 vs. Controls).
@ Total bilirubin > 2.1mg/dL is indicative of jaundice among the HIV-1 patients subgroups ($\chi 2$Mantel-Haenszel = 4.02; P = 0.05).
sGOT = serum glutamate-oxaloacetate transaminase; sGPT = serum glutamate-pyruvate transaminase ; ALK = Alkaline phosphatase;
LDH = Lactate dehydrogenase; TB = Total bilirubin.

Table 4: Regression analysis of parameters that may cause lymphocyte count reduction in HIV-1 infected patient with five-fold increase in glutamate-pyruvate transaminase activity.

Parameter	β coefficient	SEM	95% confidence Lower	Upper	P
sGOT	1.02	3.63	0.879	1.160	5.4×10^{7}
ALK	0.12	0.04	0.033	0.216	0.014
LDH	0.11	0.04	0.036	0.009	0.036
TB	0.13	0.02	0.073	0.179	8.4×10^{4}

sGOT = serum glutamate-oxaloacetate transaminase; ALK = Alkaline phosphatase; LDH = Lactate dehydrogenase
TB = Total bilirubin. P = exact probability value at 95% confidence with lower and upper values.

of 26 had total bilirubin level > 2.1mg/dl , which is indicative of jaundice (χ^2 Mantel-Haenszel = 4.02; $P = 0.05$).

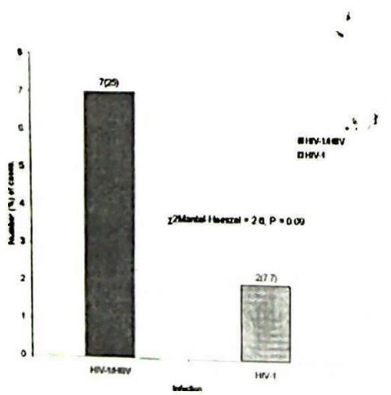

Fig. 1: *Frequency distribution of HIV-1 patients with five fold increase in glutamate-pyruvate transminase activity. Percentage are in parentheses.*

Sex distribution of HIV-1 infected patients with five fold increase in serum Glutamate pyruvate transaminase (sGPT) activity.

Biochemical indication of hepatic cytolysis (SGPT ≥ 5 x activity in normal subject) was found in 7 (25%) co-infected (4 males and 3 males) and 2 (7.7%) HIV-1 infected male patients respectively with disparity in proportion showing no significant ($P > 0.05$) (Figure 1).

Regression analysis of parameters that may cause decrease lymphocyte count in HIV-1 infected patients with five fold increase in serum Glutamate pyruvate transaminase (sGPT) activity.

In HIV-1 infected patients with biochemical evidence of hepatic cytolysis, significant associations were observed between decreased lymphocyte count and elevated levels of all the parameters measured ($P \leq 0.01$) (Table 4).

Discussion

HIV and HBV are blood borne pathogens with similar risk

factors and high propensity to co-infect humans [26,27]. Several surveillance studies have reported high morbidity and mortality in HBV infected HIV patients as a result of worsening liver pathology and rapidly declined immune responses. This scenario has undoubtedly offered new challenges to the clinical management of HIV/AIDS worldwide [28,29].

In the present study we observed 51.9% seroprevalence of HBsAg among HIV-1 infected patients that was significantly higher than 12.5% in the apparently healthy controls. The observed HBsAg antigenemia level in our HIV –1 patients is higher than 4.4% and 40% rates observed previously in Lagos among HIV seropositive women and HIV infected patients [11,12], suggesting an increment in the prevalence rate of HBV infection among HIV patients in Western Nigeria. The prevalence rate of 42.1% for HBV/HCV co-infection reported by Ola *et al* [30] further indicates changes in the pattern of HBV infection with HIV infection overwhelming HCV viremia. This may be a reflection of the increasing prevalence of HIV infection in the country [31], which calls for more aggressive control strategies and measures to curtail the spread of HIV/AIDS in Nigeria. Meanwhile, the observed 12.5% prevalence rate in our apparently healthy controls is higher than 1.9% found in children with protein energy malnutrition (PEM) [32] but lower than 23.4% found in donated blood samples in Ilorin [33], 48% among infants and toddlers [34] and 20% reported by Olatunji *et al* [35] among blood transfused sicklers in Lagos. Taken together, our findings appear to connote an improvement in the carriage of hepatitis B virus in apparently healthy Nigerians. This improvement may be as a result of improvement in the level of hepatitis B vaccination in Lagos hospitals (Hodonu *et al,* unpublished) coupled with improved handling of blood and blood contaminable materials in the state. Nigeria is one of the 116 countries enlisted in 1991 by WHO to have integrated HBV vaccination into their national Extended Programme on Immunization (EPI) programmes [36]. The clinical benefits accruable from HBV-EPI have also been reported in many Asian countries [37] and a few African countries [38].

Total lymphocyte count and CD4 T lymphocyte levels are among the parameters monitored to study disease progression and inform decision on when to commence antiretroviral therapy in HIV patients worldwide [19,39]. In the present study we observed reduced total lymphocyte count among the HIV patients with further decrease in patients seropositive for HBsAg compared to the control. Our observations agree with previous findings [40] and further reveal a deteriorating effect of HBV infection on blood lymphocyte level in HIV patients. HIV has been found to induce death of both infected and uninfected (bystanders) T cells to cause a net depletion in cell population [40]. Further profiling of the HIV patients for CD4 T lymphocyte levels and distribution majority of the patients that are due for treatment with HAART (CD4 T lymphocyte count < 200 cells/mm^3) and those that are to commence on ARV without protease inhibitors (CD4 T lymphocyte = 200 – 350cells/mm^3) [19,39] also exhibited HBsAg antigenemia. Effect management of this group of HIV patients with HAART is currently of clinical concern due to hepatotoxic potentials of protease inhibitors, whose withdrawals in some studies were found necessary for good clinical outcomes and mortality prevention [13,41]. In this study 50.6 – 104.1 % increase in the liver function parameters (sGPT, sGOT, LDH, ALK, Total bilirubin) measured were found in our HBsAg seropositive HIV patients compared to their seronegative patients. Twenty-five percent and 7.7% of these patients also exhibited five-fold increase in sGPT activity, a biochemical evidence of hepatic cytolysis demanding PIs withdrawal in disease management [39,41]. In a previous study by Saves *et al* [39], the workers observed a strong correlation between HBsAg antigenemia and hepatic cytolysis in HBV/HIV patients. Bessesen *et al* [42] also suggested that HIV/HBV co-infection could cause marked rebound in viral replication causing hepatitis flare. Although, 49.2% of our HIV/HBV patients compared to 7.7% in the HIV – infected HBsAg seronegative group had total bilirubin levels suggestive of jaundice, our data are currently weak to validate the suggestion of Besensen *et al* [42].

However, our results indicate that HBsAg seropositive HIV patients with < 200 CD4 T cells /mm^3 may be therapeutically disadvantaged with regards to HAART administration due to their higher risk of hepatic cytolysis and may compromise treatment modalities and utilization of regimens that could effectively abrogate HIV progression in this category of patients in the country.

Whereas lymphocyte count has been found to correlate poorly with CD4 T lymphocyte level [19] this study observed significant correlations between elevated liver function parameters and reduced lymphocyte counts in our HIV patients with five–fold increase in sGPT activity. Therefore, our findings imply that lymphocyte depletion may act indirectly as a marker for hepatic cytolysis risk in HIV patients with HBsAg seropositivity in Nigeria.

Based on the results of this study, it can be concluded that HBV infection complicates HIV-1 pathogenesis in patients with severe pathophysiological consequences on liver function and immune response based on total and CD4 T lymphocyte counts. Prescreening of HBsAg in Nigerian HIV-1 patients as a prelude to pharmacotherapy in order to curtail metabolic deterioration of the liver and progression to hepatic cytolysis is therefore advised.

References

1. UNAIDS. Report on global HIV/AIDS epidemic. Joint United Nations Programme on HIV/AIDS. Geneva, Switzerland. 1998.
2. Johnson, AO; Akinbami, FD; Ekambi, N;

Akinyinka, OO and Odelola, HA. Patterns of childhood hepatitis in the Nigerian African. IARC. Sci. Publ. 1984; 63: 131 – 141.

3. Harry, TO; Bajani, MD and Moses, AE. Hepatitis B virus infection among blood donors and pregnant women in Maiduguri, Nigeria. East Afr. Med. J. 1994; 71: 596 – 597.
4. Nakwagala, FN and Kaginu, MM. Hepatitis B virus and HIV infections among patients in Mulago hospital. East Afr.Med. J. 2002; 79: 68 – 72.
5. Odemuyiwa, SO; Muldersd, MN; Oyedele, OI; Ola, SO; Odaibo, GN; Olaleye, DO and Muller, CP. Phylogenetic analysis of new hepatitis B virus isolates from Nigeria supports endemicity of genotype E in West Africa. J Med. Virol. 2001; 65: 463 – 469.
6. Sirisena, ND; Njoku, MO; Idoko, JA; Isamade, E; Barau, C; Jelpe, D; Zamani, A and Otowo, S. Carriage rate of hepatitis B surface antigen (HbsAg) in urban community in Jos, Plateau State, Nigeria. Niger. Postgrad. Med. J. 2002; 9: 7 – 10
7. Ofili, AN; Asuzu, MMC and Okojie, OH. Knowledge and practice of universal precautions among nurses in central hospital, Benin-City, Edo State, Nigeria. Niger. Postgrad. Med. J. 2003; 10: 26 – 31.
8. Carman, WF. The clinical significance of surface antigen variants of hepatitis B virus. J Viral Hepat (Suppl. 1): 1992; S11-S20.
9. Baba, MM; Ajayi, BB and Ekanem, IA. Prevalence of hepatitis B surface antigen among patients suspected of liver diseases in a Nigerian hospital. Niger. Postgrad. Med. J. 2000; 7: 91 – 95.
10. Akinyinka, OO; Falade, AG; Ogunbiyi, O and Johnson, AO. Hepatocellular carcinoma in Nigerian children. Ann Trop. Paediatr. 2001; 21: 165 – 168.
11. Obi, CL; Anyiwo, CE; Nnatu, SN; Agbonlahor, DE; Esumeh, FI and Karpqas, A. A comparison of human immunodeficiency virus (HIV) seropositivity and hepatitis B surface antigenemia (HbsAg) among the same group of apparently healthy pregnant women in Lagos, Nigeria: a preliminary report. Viral Immunol. 1993; 6: 43 – 47.
12. Otuonye, NM; Olukoya, DK; Odunukwe, NN; Idigbe, EO; Udeaja, MN; Bamidele, M; Onyewuchie, JI; Oparaugu, CT; Ayelari, OS and Oyekunle, B. HIV association with conventional STDS (sexual transmitted diseases) in Lagos State, Nigeria. West. J. Med.2002; 21: 153 – 156.
13. Collier, AC; Coombs, RW; Schoenfeld, DA; Bassett, R; Baruch, A and Corey, L. Treatment of human immunodeficiency virus infection with saquinavir, zidovudine and zalcitabine. N Engl J Med.1996; 334: 1011 – 1017.
14. Ockenga, J; Tillmann, HL; Trautwein, C; Stoll, M; Manns, MP and Schmidt, RE. Hepatitis B and C in HIV-infected patients. Prevalence and prognostic value. J. Hepatol. 1997; 27: 18 –24.
15. Thibault, V; Benhamou, Y; Seguret, C; Bochet, M; Katlama, C; Bricaire, F; Opolon, P; Poynard, T and A Gut, H. Hepatitis B virus (HBV) mutations associated with lamivudine in patients coinfected with HBV and Human Immunodeficiency Virus. J Clin Microbiol. 1999; 37: 3013 – 3019.
16. Davis, GL. Interferon treatment of viral hepatitis in immunocompromised patients. Semin. Liver. Dis. 1989; .9: 267 – 272.
17. Tsen, YJ; Chang, MH; Hsu, HY; Lee, CY; Sung, JL and Chen, DS. Seroprevalence of hepatitis B virus infection in children in Tapei, 1989: five years after a mass hepatitis by vaccination programme. J Med virol. 1991; 34: 96 – 99.
18. Ayoola, EA, EY; Mohsen, ST; Mohammed, OG; Basher, SA; Margani, KH and Abdurahim, MA. The decline in hepatitis B viral infection in South-Western Saudi Arabia. Saudi Med J. 2003; 24: 991 – 995.
19. World Health Organisation. Scaling up antiretroviral therapy in resource-limited settings: Guidelines for a public health approach. Executive Summary WHO. 2002; 6 - 31
20. World Health Organisation. Hepatitis B surface antigen assays: Operational characteristics phase 1. WHO. 2001; 1 - 40
21. Dacie, JV and Lewis, SM. In: Practical haematology. 7th edition. ELBS with Churchill Livingstone. Longman Group. UK. .Ltd. 1991.
22. Bergemeyer, HU and Bent, E. Glutamatae – oxaloacetate transaminase, glutamate pyruvate transaminase. In Bergemeyer ed. Methods of Enzymatic Analysis. Academic Press New York, USA. 1963; 837 - 852.
23. Denier, CC; Brison-Lougarre, AA; Biasini, GG; Grozdea, JJ and Fournier, DD. Kinetic comparison of tissue non specific and placental human alkaline phosphatases expressed in babiculovirus infected cells: application to screening for Down's syndrome. BMC Biochem. 2003; 3: 2 – 7
24. Wroblewski,Fand Ladue,JNS. Lactic dehydrogenase activity in blood. Proc. Soc. Expt. Biol. Med. 1955; 90: 210 – 213
25. Jendrassik, L and Grof, P. Vereinfachte photometrische Methode zur Beshmung des Bilirubins. Biochem Z. 1938; 297: 81 - 89
26. Wu, N; Li, D; Zhu, B and Zou, W. Preliminary search on the co-infection of human immunodeficiency virus and hepatitis virus in intravenous drug users. Chin. Med. J. 2003; 16: 1318 – 1320
27. Kellerman, SE; Hanson, DL; McNaghten, AD and Fleming, PL. Prevalence of chronic hepatitis B

and incidence of acute hepatitis B infection in human immunodeficiency virus-infected subjects. J. Infect. Dis. 2003; 188: 571 - 577.

28. Brook, MG; Jones, K; Dale, AW and Miller, RF. Management of HIV and hepatitis B or C co-infection in 15 HIV treatment centres. Disparity between protocols and practice. Int. J. STD. AIDS. 2003; 14: 469 – 472.
29. Rockstroh, JK. Management of hepatitis B and C in HIV co-infected patients. J Acquir. Immune. Defic. Syndr. 34 (Suppl 1): 2003; S59 – S65.
30. Ola, SO; Otegbayo, JA; Odaibo, GN; Olaleye, OD and Olubuyide, OL. Serum hepatitis C virus and hepatitis B surface antigenemia in Nigerian patients with acute icteric hepatitis. West Afr. J Med. 2002; 21: 215 – 217
31. National Institute for Medical Research (NIMR) and Federal Ministry of Science and Technology. The communiqué of a 2-day national symposium on HIV/AIDS in Nigeria: Research priorities for Nigeria in the next decade and strategies for their achievement. National Institute for Medical Research (NIMR), Nigeria. Nov 10 – 11, 1997. Nigerian Journal of Medical Research. 1998; 2 (1 and 2): 35
32. Akenami, FO; Koskiniemi, M; Ekanem, EE; Bolarin, DM and Vaheri, A. Seroprevalence and coprevalence of HIV and HBsAg in Nigerian children with/without protein energy malnutrition. Acta. Trop. 1997; 64: 167 – 174.
33. Bada, AS; Olatunji, PO; Adewuyi, JO; Iseniyi, JO and Onile, BA. Hepatitis B surface antigenemia in Ilorin, Kwara State, Nigeria. Centr. Afr. J Med. 1996; 42: 139 – 141.
34. Tabor, E and Gerety, RJ. Hepatitis B virus infection in infants and toddlers in Nigeria: the need for early intervention. J Paediatr. 1979; .95: 647 – 650.
35. Olatunji, RO; Akanmu, AS; Akinsete, I and Njoku, OS. Sickle cell anaemia and the risk of transfusion transmitted human immunodeficiency and hepatitis B infections in Lagos, Nigeria. The Nigerian Journal of Medical Research. 1998; 2: 1 - 3
36. World Health Organisation. Expanded programme on immunization (EPI). Immunization schedules in the WHO African region, 1995. Wkly Epidemiol. Rec. 1996; 71: 90 – 94.
37. Faleh, FZ; Al-Jeffri, M; Ramia, S; Al-Rashed, R; Arif, M and Rezeig, M. Seroepidemiology of hepatitis B virus infection in Saudi children after a mass hepatitis B vaccination programme. J Infect. 1999; 38: 167 – 170.
38. Fortuin, M; Chotard, J; Jack, AD; Maine, NP; Mendy, M and Hall, AJ. Efficacy of hepatitis B vaccine in the Gambian expanded programme of immunization. Lancet. 1995; 341: 1129 – 1131
39. Saves, M; Raffi, F; Clevenbergh, P; Marchou, B; Waldner-Combernoux, A; Morlat, P; Moing, VL; Riviere, C; Chene, G and Leport, C. The APROCO Study Group. Hepatitis B or Hepatitis C virus infection is a risk factor for severe hepatic cytolysis after initiation of a protease inhibitor-containing antiretroviral regimen in human Immunodeficiency virus-infected patients. Antimicrobial Agents Chemother. 2000; 44: 3451 – 3455.
40. Derks, RA and Beaman, KD. Regeneration and tolerance factor prevents Bystander T cell death associated with human immunodeficiency virus infection. Clin Diagn Lab. Immunol. 2004; 11: 835 - 840
41. Brau, N; Leaf, HL; Wieczorek, RL and Margolis, DM. Severe hepatitis in three AIDS patients treated with Indinavir. Lancet. 1997; 349: 924 – 925.
42. Bessesen, M; Ivews, D; Condreay, L; Lawrence, S and Sherman, KE. Chronic active hepatitis B exacerbations in human immunodeficiency virus-infected patients following development of resistance to or withdrawal of Lamivudine. Clin. Infect. Dis. 1999; 28: 1032 – 1035.

Received: 19/07/04
Accepted: 01/08/06