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Overt and occult hepatitis B virus infection in adult Sudanese HIV patients

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SUMMARY

Objectives: Human immunodeficiency virus (HIV) infection in Sub-Saharan Africa is complicated by co-infection with hepatitis B and C viruses (HBV and HCV), which share similar transmission routes. The aims of this study were to determine the prevalence of hepatitis B surface antigen (HBsAg)-positive and HBsAg-negative HBV infection and of HCV infection among HIV-infected patients.
Methods: A cross-sectional study was conducted among treatment-naïve HIV-positive adults in Khartoum State. HBV, HCV, and HIV infections were detected using immunoassays for HBsAg, hepatitis B core antibodies (anti-HBc), hepatitis C antibodies (anti-HCV), and HIV antibodies (anti-HIV), while real-time PCR was used to measure HBV DNA.
Results: The mean age of the 358 patients was 35.2 ± 9.3 years and the male to female ratio was 1.3:1.0. The mean alanine aminotransferase (ALT) level was 10.9 ± 18.0 U/l. Evidence of 23, current or past HBV infection was detected in 62.8% of the patients. HBV DNA was detected in 96 patients (26.8%), 42 HBsAg-positive (11.7%) and 54 (15.1%) HBsAg-negative, indicating occult hepatitis B infection. Anti-HCV was detected in 1.7%.
Conclusions: Evidence of HBV infection was detected in 26.8% of HIV patients with HBsAg-negative infection, with viraemia detected in 15.1% of the patients. All HIV-infected patients should be screened carefully for HBV infection with HBsAg and anti-HBc IgG antibodies prior to starting antiretroviral therapy.

1. Introduction

Human immunodeficiency virus (HIV) infection is a serious health problem in Sub-Saharan Africa.[1] HIV infection is complicated by co-infection with other pathogens including hepatitis B and C viruses (HBV and HCV), which share similar routes of infection. Worldwide, it is estimated that over 240 million people have a chronic HBV infection[2] and 150 million have a chronic HCV infection.[3] Globally there are 34 million people infected with HIV; 69% of them reside in Sub-Saharan Africa.[1] Although the introduction of antiretroviral therapy (ART) has decreased the death rate and the incidence of AIDS-defining diseases among HIV-infected individuals, liver disease, mainly as a result of HBV or HCV co-infection, has emerged as one of the leading causes of non-AIDS-related mortality and morbidity.[4] Co-infection with HBV or HCV and HIV is characterized by more rapid progression of liver disease, including accelerated fibrosis, cirrhosis, and hepatocellular carcinoma.[5,6]

Sudan, located in north to east-central Africa, is endemic for HBV infection, with HBV exposure rates ranging from 47%[7] to 78%,[8]

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hepatitis B surface antigen (HBsAg) prevalence of 6.9%[7] to 18.7%,[9] hepatitis C antibody (anti-HCV) prevalence of 2.2%[10] to 4.8%,[11] and a prevalence of HIV among the adult population of 0.9–1.4%.[12] However, there is no clear knowledge about HBV in the setting of HIV infection, and it is important to determine this and select appropriate ART, especially as HIV patients may also develop hepatic toxicity and lactic acidosis under ART, which is based on a combination of drugs including lamivudine, stavudine, efavirenz, and nevirapine.[13]

No studies have been carried out to determine the presence of HBV DNA in the absence of HBsAg in HIV-positive Sudanese patients. Thus the aims of this study were to determine the prevalence of HBV and HCV infection and the prevalence of occult hepatitis B infection (OBI) among adult Sudanese HIV patients.

2. Materials and methods

2.1. Patients

A cross-sectional study was conducted during the period December 2010 to June 2012 in Khartoum, Sudan. All treatment-naïve adults (18 years and over) who tested positive for HIV antibodies using an indirect ELISA, attending Omdurman Medical AIDS Care Unit (OMACU) and Bashair Hospital HIV Treatment Centre in Khartoum State, were included in the study. Children younger than 18 years of age and pregnant women were excluded. The study was approved by the Ethics Committee of the Faculty of Medicine, University of Khartoum, the National Ethics Committee, Federal Ministry of Health, Sudan, and by the Human Ethics Committee of the University of the Witwatersrand, South Africa. Written informed consent was obtained from all participants before their enrolment.

2.2. Data collection

Data were collected using a specially designed questionnaire and included basic demographic information including age and gender, and potential risk factors for the transmission of infection, such as multiple sexual partners, intravenous drug use, previous history of jaundice and blood transfusion, and history of hepatitis B vaccination. All patients were classified according to the World Health Organization (WHO) clinical staging of HIV-related disease.[14]

2.3. Collection and storage of samples

Ten millilitres (10 ml) of venous blood was collected from each patient and divided into two blood collection tubes: 7 ml in ethylenediaminetetraacetic acid (EDTA) tubes and 3 ml in plain tubes for serum separation. Separated plasma and serum samples were stored at −30 °C until further analysis.

2.4. Immunoassays

The detection of anti-HIV was done using an indirect ELISA kit (Biorex; Antrium, UK). HBsAg, hepatitis B core antibody (anti-HBc) IgM and IgG, hepatitis B e antigen (HBeAg) and antibodies (anti-HBe), and anti-HCV were detected using immunoassays from Diasource (Nivelles, Belgium). The immunoassay for hepatitis D virus antibodies (anti-HDV) was from Diagnostic Automation, Inc. (Calabasas, CA, USA). The manufacturer's instructions were strictly followed for all tests.

2.5. DNA extraction and quantitative real-time PCR

HBV DNA was extracted with the Sacace Ribo Virus Kit (HBV Real-TM Quant; Sacace Biotechnologies, Como, Italy), as recommended by the manufacturer. An internal control (HBV IC) with known viral concentration (specific for each lot) was used to monitor the amplification and exclude any loss of genomic DNA or PCR inhibition and to enable the precise calculation of the HBV viral load in the samples. The extracted DNA was used immediately for PCR. Real-time PCR was done using the HBV Real-TM Quant Kit (Sacace Biotechnologies, Como, Italy). The extracted HBV DNA (12.5 μl) was added to an equal volume of the reaction mixture provided in the kit. With each run, three known standards of the HBV DNA (QS1 HBV, QS2 HBV, and QS3 HBV) and the three internal control standards (QS1 IC, QS2 IC, and QS3 IC) were used to calculate the viral copy numbers. Real-time PCR was done in accordance with the manufacturer's instructions, as follows: stage 1, 95 °C for 15 min; stage 2, a two-temperature cycle of 95 °C for 20 s and 60 °C for 40 s, which was repeated 42 times. Cy3 dye was used for the sample and FAM for the internal control. The signals from these dyes were calculated and presented as numbers. The sensitivity of the assay is approximately 20 genome copies/ml, which is equivalent to 12 IU/ml (1.1 $\log_{10}$ IU/ml), conversion factor = 1.7.

2.6. Statistical analysis

The statistical analysis was done using SPSS software program version 20 (SPSS Inc., Chicago, IL, USA); the Chi-square test using the *t*-statistic was performed, and significance was set at $p < 0.05$.

3. Results

A total of 358 HIV-infected patients were enrolled in the study. Their mean (± standard deviation) age was 35.2 ± 9.3 years; the male to female ratio was 1.3:1.0. The mean alanine aminotransferase (ALT) level of the study population was 10.9 ± 18.0 U/l. Demographic data of the study population as a whole and stratified according to anti-HBc IgG, as well as risk factors for acquiring HBV infection are shown in Table 1.

3.1. HBV serology

Of the 358 patients, 225 (62.8%) were anti-HBc IgG-positive, indicating previous exposure to HBV infection. The mean age and ALT level of this subpopulation were 35.3 ± 9.4 years and 11.0 ± 20.9 U/l, respectively, and these values did not differ from those of the population as a whole. Only four patients (1.1%) reported being vaccinated against HBV infection, however anti-hepatitis B surface antibodies (anti-HBs) were not tested. Male gender and intravenous drug use were found to be risk factors for positivity for HBV markers ($p < 0.05$) (Table 1).

3.2. HBsAg-positive patients

Forty-two patients (11.7%) were HBsAg-positive. The mean age and ALT level of this group were 34.7 ± 8.7 years and 9.8 ± 7.2 U/l, respectively, and these values did not differ from those of the population of 225 patients who were anti-HBc IgG-positive. Eight patients (19.0%) were HBeAg-positive and anti-HBe-negative, 24 patients (57.1%) were HBeAg-negative and anti-HBe-positive, and 10 patients (23.8%) were negative for both HBeAg and anti-HBe. All patients were negative for anti-HDV.

3.3. Viral loads

HBV DNA was detected by PCR in 96 patients (26.8%); all were anti-HBc IgG-positive. Demographic data for patients with detectable HBV DNA on PCR and risk factors for acquisition are shown in Table 2. Risk factors associated with HBV DNA positivity included blood transfusion, previous dental procedures, history of

Table 1
Demographic data and risk factors for HBV exposure of 358 HIV-positive Sudanese adults

Variable		Number of patients tested ($N = 358$)	Number positive for anti-HBc IgG ($n = 225$)	Number negative for anti-HBc IgG ($n = 133$)	p-Value
Gender	Male	203 (56.7%)	139 (61.8%)	64 (48.1%)	0.012[a]
	Female	155 (43.3%)	86 (38.2%)	69 (51.9%)	
Marital status	Married	214 (59.8%)	136 (60.4%)	78 (58.6%)	0.896
	Single	82 (22.9%)	50 (22.2%)	32 (24.1%)	
	Divorced/widowed	62 (17.3%)	39 (18.8%)	23 (17.3%)	
Multiple sexual partners	Yes	144 (40.2%)	95 (42.2%)	49 (36.8%)	0.316
	No	214 (59.8%)	130 (57.8%)	84 (63.2%)	
Surgery	Yes	65 (18.2%)	37 (16.4%)	28 (21%)	0.274
	No	293 (81.8%)	188 (83.6%)	105 (79%)	
Blood transfusion	Yes	38 (10.6%)	20 (8.9%)	18 (13.5%)	0.168
	No	320 (89.4%)	205 (91.1%)	115 (86.5%)	
Dental procedures	Yes	92 (25.7%)	54 (24.0%)	38 (28.6%)	0.339
	No	266 (74.3%)	171 (76.0%)	95 (71.4%)	
Intravenous drug use	Yes	5 (1.4%)	1 (0.4%)	4 (3%)	0.046[a]
	No	353 (98.6%)	224 (99.6%)	129 (97%)	
History of jaundice	Yes	111 (31.0%)	78 (34.7%)	33 (24.8%)	0.51
	No	247 (69.0%)	147 (65.3%)	100 (75.2%)	
HBV vaccination	Yes	4 (1.1%)	2 (0.9%)	2 (1.5%)	0.593
	No	354 (98.9%)	223 (99.1%)	131 (98.5%)	
WHO classification	Stage 1 and 2	126 (35.2%)	66 (29.3%)	60 (45.1%)	0.168
	Stage 3 and 4	232 (64.8%)	159 (70.7%)	73 (54.9%)	

HBV, hepatitis B virus; anti-HBc, hepatitis B core antibodies; WHO, World Health Organization.
[a] Significant risk factor.

Table 2
Comparison between patients who were HBV DNA-positive and patients who were HBV DNA-negative on PCR, among 358 HIV-infected patients

Variable		Number of patients tested ($N = 358$)	HBV DNA-positive ($n = 96$)	HBV DNA-negative ($n = 262$)	p-Value
Gender	Male	203 (56.7%)	59 (61.5%)	144 (55%)	0.272
	Female	155 (43.3%)	37 (38.5%)	118 (45%)	
Marital status	Married	214 (59.8%)	62 (64.6%)	152 (58%)	0.342
	Single	82 (22.9%)	24 (25%)	58 (22.1%)	
	Divorced/widowed	62 (17.3%)	10 (10.4%)	52 (19.8%)	
Multiple sexual partners	Yes	144 (40.2%)	42 (43.8%)	102 (38.9%)	0.410
	No	214 (59.8%)	54 (56.2%)	160 (61.1%)	
History of surgery	Yes	65 (18.2%)	17 (17.7%)	48 (18.3%)	0.894
	No	293 (81.8%)	79 (82.3%)	214 (81.7%)	
Blood transfusion	Yes	38 (10.6%)	5 (5.2%)	33 (12.6%)	0.044[a]
	No	320 (89.4%)	91 (94.8%)	229 (87.4)	
Dental procedures	Yes	92 (25.7%)	16 (16.7%)	76 (29%)	0.018[a]
	No	266 (74.3%)	80 (83.3%)	186 (71%)	
Intravenous drug use	Yes	5 (1.4%)	0	5 (1.9%)	0.173
	No	353 (98.6%)	96 (100%)	257 (98.1%)	
History of jaundice	Yes	111 (31.0%)	38 (39.6%)	73 (27.9%)	0.034[a]
	No	247 (69.0%)	58 (60.4%)	189 (72.1%)	
HBV vaccination	Yes	4 (1.1%)	1 (1%)	3 (1.1%)	0.934
	No	354 (98.9%)	95 (99%)	259 (98.9%)	
WHO classification	Stage 1 and 2	126 (35.2%)	23 (24%)	103 (39.3%)	0.037[a]
	Stage 3 and 4	232 (64.8%)	73 (76%)	159 (60.7%)	

HBV, hepatitis B virus; WHO, World Health Organization.
[a] Significant risk factor.

jaundice, and WHO clinical stages 3 and 4 ($p < 0.05$) (Table 2). Forty-two patients (11.7%) were HBsAg-positive and 54 patients (15.1%) were HBsAg-negative, indicating occult infection (Table 3). The only risk factor that differentiated HBsAg-positive and HBsAg-negative infections was a history of jaundice (Table 3). Moreover, the overall median (interquartile range) viral load was 2.9 (1.7–4.1) $\log_{10}$ IU/ml, with HBsAg-positive individuals having significantly higher viral loads of 3.2 (3.3–6.6) $\log_{10}$ IU/ml compared to HBsAg-negative patients with viral loads of 1.7 (1.3–2.7) $\log_{10}$ IU/ml. Eight HBsAg-positive, HBeAg-positive, anti-HBe-negative patients had viral loads of 5.6 $\log_{10}$ IU/ml, 24 HBsAg-positive, HBeAg-negative, anti-HBe-positive patients had viral loads of 3.8 $\log_{10}$ IU/ml, and 10 HBsAg-positive, HBeAg/anti-HBe-negative had viral loads of 3.9 $\log_{10}$ IU/ml. Thirty-five (9.8%) HBsAg-negative patients had a viral load < 2.3 $\log_{10}$ IU/ml (< 200 IU/ml) and 19 (5.3%) had viral loads >2.3 $\log_{10}$ IU/ml (>200 IU/ml). One patient was reactive in both anti-HBc IgG and anti-HBc IgM; the patient was HBsAg-negative with normal ALT and had detectable HBV DNA on PCR indicating a recent HBV infection or an HBV reactivation in a patient with OBI.

3.4. HCV infection

Six patients (1.7%), five of whom were female, tested positive for anti-HCV antibodies; their mean age was 34.3 ± 12.9 years and mean ALT level was 6.0 ± 3.8 U/l. None of them was an intravenous drug user and there were no significant risk factors for HCV infection. Five of these patients were anti-HBc IgG-positive, with two having detectable HBV DNA on PCR. One patient was HBsAg-positive, while the second patient was HBsAg-negative, indicating occult HBV infection.

Table 3
Comparison between HBsAg-positive and HBsAg-negative patients among 96 HIV-infected patients with detectable HBV DNA on PCR

Variable		Number of patients tested ($n=96$)	HBsAg-positive ($n=42$)	HBsAg-negative ($n=54$)	p-Value
Gender	Male	59 (61.5%)	24 (57.1%)	35 (64.8%)	0.444
	Female	37 (38.5%)	18 (42.9%)	19 (35.2%)	
Marital status	Married	62 (64.6%)	28 (66.7%)	34 (63%)	0.819
	Single	24 (25%)	11 (26.2%)	13 (24%)	
	Divorced/widowed	10 (10.4%)	3 (7.2%)	7 (13%)	
Multiple sexual partners	Yes	42 (43.8%)	18 (42.9%)	24 (44.4%)	0.876
	No	54 (56.2%)	24 (57.1%)	30 (55.6%)	
Surgery	Yes	17 (17.7%)	8 (19%)	9 (16.7%)	0.762
	No	79 (82.3%)	34 (81%)	45 (83.3%)	
Blood transfusion	Yes	5 (5.2%)	3 (7.1%)	2 (3.7%)	0.452
	No	91 (94.8%)	39 (92.9%)	52 (96.3%)	
Dental procedures	Yes	16 (16.7%)	9 (21.4%)	7 (13%)	0.270
	No	80 (83.3%)	33 (78.6%)	47 (87%)	
Intravenous drug use	Yes	0	0	0	-
	No	96 (100%)	42 (100%)	54 (100%)	
History of jaundice	Yes	38 (39.6%)	23 (54.8%)	15 (27.8%)	0.007[a]
	No	58 (60.4%)	19 (45.2%)	39 (72.2%)	
HBV vaccination	Yes	1 (1%)	1 (2.4%)	0	0.254
	No	95 (99%)	41 (97.6%)	54 (100%)	
WHO classification	Stage 1 and 2	23 (24%)	11 (26.2%)	12 (22.2%)	0.947
	Stage 3 and 4	73 (76%)	31 (73.8%)	42 (77.8%)	

HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; WHO, World Health Organization.
[a] Significant risk factor.

4. Discussion

The 62.8% exposure rate to HBV infection and prevalence of HBsAg of 11.7% detected in the HIV-positive Sudanese patients in the present study did not differ from HBV exposure rates found previously in HIV-negative patients, with rates ranging from 47%[7] to 78%[8] and the prevalence of HBsAg from 6.9%[7] to 18.7%.[9] Although HBc IgG-positive, HBsAg-positive serology can also be observed during late acute HBV infection, in Sub-Saharan Africa this would be unlikely given the epidemiology of HBV infection in the region, thus we believe that the 11.7% of patients with positive HBsAg had chronic HBV infection.

The prevalence of HCV antibodies of 1.7% was also similar to HCV prevalence rates in HIV-negative patients of 2.2%[10] to 4.8%[11] in previous studies. Similar exposure rates to HBV and HCV among HIV-positive and HIV-negative individuals in Sub-Saharan African countries have been reported.[15] In Ethiopia, in HIV-positive patients, the exposure to HBV was reported to be 47.5% and prevalence of HBsAg 3.9%, compared to 41.9% exposure to HBV and 5.4% HBsAg positivity in HIV-negative patients.[16] In Uganda, exposure to HBV and prevalence of HBsAg were reported to be 59% and 8.3%, respectively, in HIV-positive patients, compared to 51.9% and 10.4%, respectively, in HIV-negative patients.[17] A study in Mozambique in HIV-positive patients revealed exposure to HBV to be 69.6%, HBsAg 10.6%, and HCV antibodies 1.4%, compared to 62.5%, 9.0%, and 1.5%, respectively, in HIV-negative individuals.[18]

The similar frequencies of HBV infection in HIV-infected and HIV-uninfected individuals in regions of high HBV endemicity such as in Sub-Saharan Africa, suggest that the mode and timing of transmission of HBV are generally independent to those of HIV. In Sub-Saharan Africa, most HBV infections occur in childhood either perinatally or more often through horizontal transmission such as close contact within households, medical procedures using non-sterile equipment, and through traditional medical practices long before exposure to HIV.[19,20] This is in contrast to countries with low HBV endemicity such as the USA, Europe, and Australia, where HBV is frequently transmitted sexually or via intravenous drug use,[21] and may often be co-transmitted with HIV. Here, although the HBV prevalence in HIV-positive patients may be similar to that found in high endemicity regions (with Soriano et al.,[22] for example, reporting a 7.1% HBsAg positivity), it is relatively higher when compared to HIV-negative patients. Studies have also shown that following acute HBV infection, HIV patients are more likely to develop chronic infection than non-HIV patients and this may explain why HIV patients have a higher rate of chronic HBV infection in low endemicity regions.[21,23]

The present study showed a slightly higher prevalence of HBeAg (19%) among HBV/HIV co-infected patients when compared with our previous report on the prevalence of HBeAg among HIV-negative patients, in which only 12% were HBeAg-positive.[24] This, however, was not significantly different. A similar trend was reported from Zambia, where HBeAg was positive in 25% of HIV-positive patients compared to 8.5% among HIV-negative patients.[25] It is possible that HIV, by causing immunosuppression, may delay or prevent seroconversion to the inactive carrier phase in patients with chronic HBV infection.[21] It has been demonstrated that efficient restoration of immunity after the introduction of ART leads to high rates of seroconversion, especially in patients in the immunoactive phase of HBV infection.[26] However, there is also the possibility that the HIV co-infected patients, who were significantly younger than the mono-infected liver disease patients, did not have enough time to seroconvert or were recently infected with HBV. Patients infected with HBV/HIV had higher HBV viral loads compared to HBV mono-infected patients ($p < 0.05$)[24] and also lower ALT levels, within the normal range, compared to the patients infected with HBV only ($p < 0.05$).[24] The relatively low ALT levels have also been demonstrated in studies from South Africa, where there was no significant difference in ALT levels among HIV patients serologically positive or negative for HBV infection.[27] The immunosuppression caused by the HIV infection is responsible for these differences[28] and can account for the low rate of clinical manifestations of liver disease in the study population.

In addition to this risk, ART may cause further liver damage, either through direct toxicity or through idiosyncratic reactions.[29] HBV/HIV co-infection is known to increase the risk of hepatotoxicity from ART when compared to other HIV patients without HBV or HCV co-infection.[13,30] In Sub-Saharan Africa, however, this risk is not entirely attributable to ART but may also be a result of elevated levels of HBV DNA in the serum.[31]

HIV infection is also a known risk factor for the development of OBI;[32] this is characterized by two main criteria: the absence of HBsAg and low viral replication. This has been reported in several

clinical contexts, the most common being chronic carriage with HBsAg too low to be detected using current serological tests and the presence of anti-HBc IgG as the only serological evidence of exposure to the virus, referred to as 'isolated anti-HBc'.[33] In this study, 96 patients (26.8%) had detectable HBV DNA on PCR, all of them were HBc IgG-positive, 11.7% were HBsAg-positive, and 54 patients (15.1%) were HBsAg-negative indicating 'occult' infection. OBI was further defined by the Taormina Consensus Conference in 2008 as the "presence of HBV DNA in the liver (with detectable or undetectable HBV DNA in serum) of individuals testing HBsAg-negative with currently available assays and a serum HBV DNA of <2.3 $\log_{10}$ IU/ml (<200 IU/ml) when detectable".[34] In this study, 35 (9.8%) HBsAg-negative patients had a viral load <2.3 $\log_{10}$ IU/ml (<200 IU/ml) and 19 (5.3%) had viral loads >2.3 $\log_{10}$ IU/ml (>200 IU/ml). Some individuals with chronic HBV infection may be HBsAg-negative but have HBV DNA levels comparable to patients with positive HBsAg (>2.3 $\log_{10}$ IU/ml; >200 IU/ml). These individuals may be infected with HBV variants with S gene escape mutants, which do not express HBsAg on serology, as pre-S mutations may prevent HBsAg secretion.[35] In many HIV patients with OBI, the viral load may be higher than suggested by the Taormina conference, which did not deal explicitly with HIV co-infection. A similar study from South Africa has reported positive HBV DNA in 23.8% of HIV patients; 8.7% were HBsAg-positive and 15.7% were HBsAg-negative, with a viral load of <2.3 $\log_{10}$ IU/ml (<200 IU/ml) detected in 6.7% of HBsAg-negative patients.[27] OBI has been demonstrated in 21.3% of HIV-infected patients from the Cote d'Ivoire[36] and in 10.5% of cases from the USA.[37]

While HIV is treated with a combination of three drugs, which include lamivudine, stavudine, efavirenz, and nevirapine,[13] only lamivudine is active against HBV. While earlier reports have suggested rates of HBV resistance of up to 20% per year with lamivudine monotherapy in HBV/HIV co-infected patients,[38] reports from Sub-Saharan Africa have suggested that although lamivudine resistance can occur in the setting of incomplete HBV suppression, this is infrequently observed among HBV/HIV co-infected patients who are HBeAg-negative with low baseline HBV levels.[39] In 2010, the WHO recommended the introduction of ART that includes tenofovir in combination with lamivudine or emtricitabine as first-line therapy in all HBV/HIV co-infected patients needing treatment in order to reduce the development of HBV drug resistance.[40]

Interestingly, in this study, out of 96 patients with detectable HBV DNA, HIV/HBV/HCV triple infection was detected in two patients (2%); none had HIV/HBV/HDV triple infection. This is in contrast to another study in which HIV/HBV/HCV was detected in 6% and HIV/HBV/HDV was detected in 4% of patients.[41] It is generally thought that HBV replication is significantly reduced in HCV and HDV triple infections. The low HCV triple infection in this study could be explained by the low prevalence of HCV of only 2.2–4.8% found in Sudan.[10,11] However, the absence of HDV triple infection is difficult to explain considering that HDV antibodies were detected in 9.0% of patients with chronic HBV mono-infection.[42]

Limitations of the present study include not testing for anti-HBs to determine immunity to HBV in HBsAg-negative patients, not determining CD4 counts or HIV viral loads, and not performing nucleic acid testing for HCV, liver fibrosis assessment, or multivariable analysis.

In conclusion, in Sudan exposure rates to HBV infection and the prevalence of HBsAg and HCV antibodies among adult HIV-infected patients were found to be similar to those reported for HIV-negative patients in previous studies. HBsAg-negative HBV infection was detected in 15.1% of patients. The prevention of HBV and HIV through education and vaccination against HBV is essential in our setting. All HIV-infected patients should be screened carefully for HBV infection with HBsAg and anti-HBc IgG antibodies prior to starting ART, which should include tenofovir and lamivudine (or emtricitabine) as first-line therapy. Although PCR testing would detect OBI, which would be missed by serological testing alone, it is too expensive to include as a routine screening test in Sudan. Considering that nucleic acid testing can detect both overt and occult HBV infection, cost-effective alternatives could include PCR testing only in patients with clinical or laboratory evidence of active liver disease, or the development of a cheap, rapid test for HBV DNA detection.

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