International Journal of TROPICAL DISEASE & Health
8(2): 75-78, 2015, Article no.IJTDH.2015.079
ISSN: 2278–1005

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Prevalence of Anti-HBc in HIV Patients on ART in Ekiti, Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Author OOO did the study design and wrote the protocol. Authors OAS collected the samples, wrote the manuscript and did stastical analysis. Authors OJ and OG did laboratory experiment. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJTDH/2015/17400
Editor(s):
(1) William Ebomoyi, Department of Health Studies, College of Health Sciences, Chicago State University, USA.
Reviewers:
(1) Anonymous, Regents University Augusta, USA.
(2) Mathew Folaranmi Olaniyan, Department of Medical Laboratory Science, Achievers University, Nigeria.
(3) Anonymous, University of Tromsø, Norway.
Complete Peer review History: http://www.sciencedomain.org/review-history.php?iid=1133&id=19&aid=9073

Case Study

Received 12th March 2015
Accepted 14th April 2015
Published 2nd May 2015

ABSTRACT

Background: In HIV –infected individuals, anti-HBc which is an indicator of occult hepatitis B is a common phenomenon
Aim: The aim of this study is to determine the sero-epidemiology and associated risk factors among HIV-infected individuals in Ikole Ekiti, Nigeria.
Study Population and Duration: HIV/AIDS individuals visiting Institute of Human Virology of Nigeria, Specialist Clinic, Ikole Ekiti between November 2012 and April 2013 were included in this study.
Methodology: One hundred and eighty eight HIV samples were tested for anti-Hepatitis B core antibody by Enzyme Linked Immunosorbent Assay (ELISA) {ANTICORASE MB-96 [TMB] from General Biological Corporation}. Data were analysed using software within SPSS packages.
Results: Twenty two (11.7%) of 188 HIV-infected patients had isolated anti-HBc. Co-infection of HIV-HBV in males (3.23%) differed significantly from that of females (8.51%). Highest coinfection

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(9.57%) was found among adult age group of 20 - 55, while no co-infection was found in teenager age group (13-19 years). Based on occupation, long distance drivers had highest coinfection of 8 (4.26%) while student had lowest prevalence of 2 (1.06%). Isolated anti-HBc among HIV individuals was found in 22 (11.7%) which was significantly high.

Conclusion: Since presence of Anti-HBc has been proved to be an indicator of occult hepatitis B which can cause liver cirrhosis and later lead to hepatocellular carcinoma therefore all patients attending HIV-clinic should also be tested for anti-HBc before commencement of Highly active antiretroviral therapy (HAART).

Keywords: HBV; HIV; Anti-HBc coinfection.

1. INTRODUCTION

Despite an uncommon serological pattern in the general population, isolated anti-HBc, defined by positivity of anti-HBc marker in absence of both HBV surface antigen (HBsAg) and anti-HBV surface antibody (anti-HBs) markers, is usually seen in HIV-positive patients [1]. Nevertheless, the clinical importance and implications of isolated anti-HBc detection remain not well defined in the HIV-positive population. Only infected hepatocyte harbored Hepatitis B core antigen which stimulate the production of antibody which will be detected in the serum [2]. Anti-HBc is very essential in HBV diagnosis because of its presence throughout the course of infection and cannot be found in immunized individuals. Immunoglobulin IgM and IgG are found in serum during acute (not more than 6 month) and chronic infection [2]. It does not confer protection against HBV. Anti-HBc antibodies are markers of acute, chronic, or resolved HBV infection and remain detectable for life. These can be present in the absence of both HBsAg and anti-HBs antibodies, during the convalescent period following acute hepatitis B before the appearance of anti-HBs antibodies, or in patients who have resolved infections but lost detectable anti-HBs antibodies. Anti-HBc antibodies are, therefore, detected in anyone who has been infected with HBV [3].

Anti-HBc is the first antibody to developin less than 8 weeks after exposure to HBV. The anti-HBc is present in the serum throughout life time of infected individuals and total antibody appear after 6 months [4]. In patients who cleared acute HBV infection IgM anti-HBc is not usually absent after 6 months, otherwise the IgM anti-HBc is present after many years of infection [5].

2. MATERIALS AND METHODS

The study subjects were confirmed HIV patients attending IHVN Clinic in Nigeria. The patients were mainly Yoruba and almost all of them were on Antiretroviral Treatment (ART) in thehospital. Ethical approval was obtained for from Ministry of Health, Ekiti State. Informed consent was obtained from each study subject before administration of questionnaire. From 200 HIV positive samples collected from IHVN Clinic in Nigeria, 188 HIV samples were randomly selected for the project. 4ml of venous blood was collected from each participant aseptically by venepuncture from the cubital fossa into sterile plain bottles containers using a standard procedure. The blood was allowed to clot and centrifuged at 3000 rpm for 5 minutes to separate the serum. The serums so extracted were stored in plain bottle at –20ºC until tested.

Required number of strips in the folder was placed and wells for standard and samples were identified carefully. Samples were diluted in ratio 1:101 by dispensing 1ml of sample diluents into a disposable tube and then 10µl sample; both were mixed on vortex before use. A1 + B1 were left empty for blanking purpose. 100 µl of calibrators, dissolved control serum and diluted samples were pipetted in duplicate respectively. Microplate was incubated for 60 mins at 37ºC. After the 1st incubation, microwells were washed. In all the wells except A1+B1, 100 µl of enzyme conjugate was pipetted.

Microplate was incubated at 37ºC for 60mins. After the 2nd incubation, microwells were washed. 100µl of chromogen/substrate was pipetted into all the wells (A1+B1 inclusive).Microplate protected from light was incubated at room temperature (18-24ºC for 20 mins. 100 µl of sulphuric acid was pipetted into all the wells to block the enzymatic reaction. Addition of the stop solution turned the positive control and positive samples from blue to yellow. Colour intensity of the solution in each well was measured using 450 nm filter reading.

3. RESULTS

A total of 188 HIV infected individuals comprising of 43 males and 145 females with mean age 37 yrs (7-75) years were investigated for Anti HBc IgM. Table 1 shows demographic characteristic of study population. Twenty two (11.7%) of 188 HIV individuals patients were positive for Anti HBc IgM with 3.19% and 8.51 seroprevalence rate among the males and female respectively. The mean age of the Anti HBc IgM positive patients was 36years. Highest coinfection rate of 9.57% was found in 20-55 years age group (9.57%), followed by 1-12 (1.06%), >56 (1.06) and 13-19 (0.0%). In relation to marital status, single had the highest rate of co-infection (9.57%), while married had 2.13%. Table 2 shows Anti HBc IgM HIV frequency among occupational groups. The average CD4 count of AntiHBc positive and AntiHBc negative HIV patients was 378.5 cell/mm^3 and 470 cellmm3 respectively. The overall CD4 count of the study population was 459.8 cell/mm^3. Highest co-infection rate was found among long distance drivers (4.26%) followed by civil servants (3.19%) and housewives (2.13%).

Table 1. Showing demographic characteristic of studied population

Age group	Anti-HBc IgM positive	Anti-HBc IgM negative
	Number (%)	Number (%)
1-12	2 (1.06%)	4 (2.13%)
13-19	0	20 (10.6%)
20-55	18 (9.57%)	122(64.9%)
>56	2 (1.06%)	20(10.6%)
Mean (Range)	36yrs (9.5-60)	37 yrs(7-75)
Male	6 (3.19%)	36(19.1%)
Female	16(8.51)	130(69.1%)
Marital status		
Married	4(2.13%)	142(75.53%)
Single never married	18(9.57%)	24(12.8%)
CD4 count		
<200	4	14
200-499	6	62
>500	12	90
Mean (cell/mm^3	378.5	470

4. DISCUSSION

This study showed Anti HBc IgM prevalence rate of 11.7% in patient on ART in Ikole Ekiti which is an agreement with Sirisena et al. [6] but in contrary to 15% reported by Baba et al. [5,4,6]. Older age and CD4 counts were independently associated with presence of isolated anti-HBc. Similar to these findings; two studies also found that decreased CD4 values were linked with presence of anti-HBc in patients on ART [7]. Patients in most of the other studies had higher CD4 counts (median CD4 counts of 200 cells/mm^3). Few studies have addressed the association between age and isolated anti-HBc. In an unselected population in Germany, highest co-infection of isolated anti-HBc was found among aged of 71–80 years (22.7%) and lowest in people aged 1–30 years (9.4%) [8]. Unlike our observations, one study reported HIV-positive individuals with isolated anti-HBc were older than those tested negative for all three HBV markers (mean 36.8 vs. 34.3 years, P=0.026) [9]. Because anti-HBs titers decline with age, it is not unexpected to observe that older age was associated with isolated anti-HBc. This further suggests factors associated with declining immunity, such as old age or low CD4 counts; also contribute to the presence of isolated anti-HBc. A study reported that a significant proportion of carriers go on to develop chronic hepatitis, and some may even progress to hepatocellular carcinoma but did not differentiate carriers of AntiHBc IgM from those with active infection [10].

Table 2. Anti HBc IgM-HIV frequency among occupational groups

Occupation	No of positive	Percentage positive
Businessmen	2	1.06
Civil servants	6	3.19
Drivers	8	4.26
Housewives	4	2.13
Students	2	1.06

5. CONCLUSION AND RECOMMENDA-TION

In conclusion, isolated anti-HBc was present in 11.7% of HIV-positive patients. Presence of isolated anti-HBc was associated with increase age and severe immunosuppression with CD4; suggesting compromised immunity may play a role. Co-infection rate between males (3.19%) and females (8.51%) was not significant (p>0.5), which is in not in agreement with the findings of Baba et al. [5].

Highest rate of AntiHBc IgM positivity of age distribution of 9.57% was found in age group 20-55 years. There was relatively AntiHBc IgM positivity in the age groups 0 -12 years and >56 years, but no cases was seen in 13 - 19 years age group. This is strange since a larger percentage of infection occur at early age [11]. Result of this finding may be as a result of low number of children represented in the group. High rate of co-infection was recorded in those who were single never married than among the married which may be due to infidelity and sexual workers patronage. Long distance drivers had higher co-infection rate which may be as a result of high risk behaviours found in this group [11]. Finally, HIV- anti-HBc coinfection rate is high and therefore there is need to screen all HIV infected individuals for Anti- HBc before Antiretroviral therapy.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history.php?iid=1133&id=19&aid=9073