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Original Article

QUANTIFICATION OF HEPATITIS B AND HEPATITIS C VIRAL LOAD BY REAL-TIME PCR AND ASSESSMENT OF COINFECTION OF HBV AND HCV AMONG HIV-POSITIVE PATIENTS IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Objective: To assess the prevalence of coinfection of HIV-HBV and HIV-HCV and to estimate the viral load of HBV and HCV in PLWH.

Methods: A retrospective study was conducted from November 2021 to October 2022 among patients attending ICTC. 5 ml of blood sample collected aseptically was tested for HIV, HBV and HCV using rapid immunochromatographic tests, ELISA and viral load estimated by Real-time PCR.

Results: Out of 5087 samples tested for HIV, 666 samples (13.09%) were found to be positive. Prevalence of HIV-HBV and HIV-HCV coinfection was 15.6%(104 cases) and 1.5% (10 cases), respectively. Out of which, males were predominant (62.28%). This is clinically significant with a p-value of * 0.05. HIV-HBV and HIV-HCV coinfections were predominant in 41-50 y age group. Among104 HIV-HBV coinfected, viral load at the time of diagnosis is 'below detection level' in 25(24.04%), <250 in 15(14.42%), 251-500 in 9(8.65 %), 501-1000 in 13(12.5 %), 1001-10,000 in 23(22 %) and>10, 000 copies/ml in 19(18.26%). In 10 HIV-HCV coinfected cases, the viral load is 'below detection level' in 2(20%), <250 in 1(10%), 251-500 in 1(10 %), 501-1000 in 1(10 %), 1001-10,000 in 4(40%) and>10,001-100,000 copies/ml in 1(10%).

Conclusion: Monitoring the viral load in HBV or HCV infected at the time of diagnosis of HIV or testing for protective levels of antibodies post-vaccination in uninfected people will help in limiting the progression of chronic HBV or HCV to cirrhosis, end-stage liver disease or hepatocellular carcinoma.

Keywords: Co-infection, HIV, HBV, HCV, Viral load

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INTRODUCTION

Human immunodeficiency viruses (HIV-1/2), Hepatitis B virus (HBV) and Hepatitis C virus (HCV) are important blood-borne viruses [1]. HBV and/or HCV co-infection is common among people living with HIV infection (PLWH) because of common modes of transmission like sexual intercourse, parenteral routes such as blood transfusion, sharing of needles, needle-stick injury, organ transplantation [2]. HIV, hepatitis B virus (HBV), hepatitis C virus (HCV) infections are associated with systemic manifestations and grave consequences [3]. Both HBV and HCV show hepato-tropism and individually each of these infections can cause high mortality and high morbidity [3]. However, when they are co-infected with HIV, the natural course of HBV and HCV infection is altered and it facilitates earlier progression of the disease to liver failure, cirrhosis, hepatocellular carcinoma or death [3-5]. This study, therefore, aims to understand the prevalence of HBV and HCV co-infection in PLWH and the need for timely screening, early diagnosis and treatment of these infections.

MATERIALS AND METHODS

The study was a retrospective study conducted over a period of one year (from November 2021 to October 2022) among patients attending ICTC, Department of Microbiology, King George Hospital, Visakhapatnam, Andhra Pradesh. All attendees were screened for HIV, Hepatitis B and Hepatitis C infection after taking informed consent.5 ml of blood sample was collected aseptically and tested for HIV according to NACO guidelines 2021 with three kits HIV 1+2Immunodot test kit (COMBAIDS-RS Advantage-ST), STANDARD Q HIV 1/2 Ab Test and HIV-1/2 Flowthrough Test. The samples were simultaneously tested for HBV and HCV using rapid immunochromatographic tests (HBsAg Test Kit-OZONE BIOMEDICALS, STANDARD HCV Ab Rapid Test) and further confirmed by ELISA. Viral loads (HIV RNA, HBV DNA, HCV RNA) were quantified by Real-time PCR amplification. All the tests were carried out strictly as per manufacturer's instructions.

RESULTS

Total 5087 patient samples were tested for HIV as per NACO guidelines 2021 in the ICTC department, King George Hospital, Visakhapatnam, during the study period of one year from November 2021 to October 2022. Out of these 5087 samples tested for HIV, 666 samples (13.09%) were found to be seropositive and 4421 (86.90%) were seronegative. The Sero-prevalence of HIV in ICTC attendees is shown in table 1.

Table 1: Sero-prevalence of HIV in ICTC attendees

Total samples tested	Total sero positives	Total sero negatives
5087	666 (13.09%)	4421 (86.90%)

Among these 666 HIV Sero positives, prevalence of HIV-HBV and HIV-HCV coinfection was 15.6% (104 cases) and 1.5% (10 cases), respectively. Prevalence of HIV-HBV and HIV-HCV co-infection is shown in table 2.

Table 2: Prevalence of HIV-HBV and HIV-HCV co-infection

Total HIV seropositives	HIV and HBV coinfection	HIV and HCV coinfection
666	104 (15.6%)	10 (1.5%)

Out of these 114 total co-infected cases, males were 71(62.28%), females were 41(35.96 %) and transgenders were 2 (1.75%). This is clinically significant with a p value of *0.05. Gender-wise distribution of study population is shown in fig. 1 and sex-wise distribution of HIV-HBV and HIV-HCV coinfection in table 3.

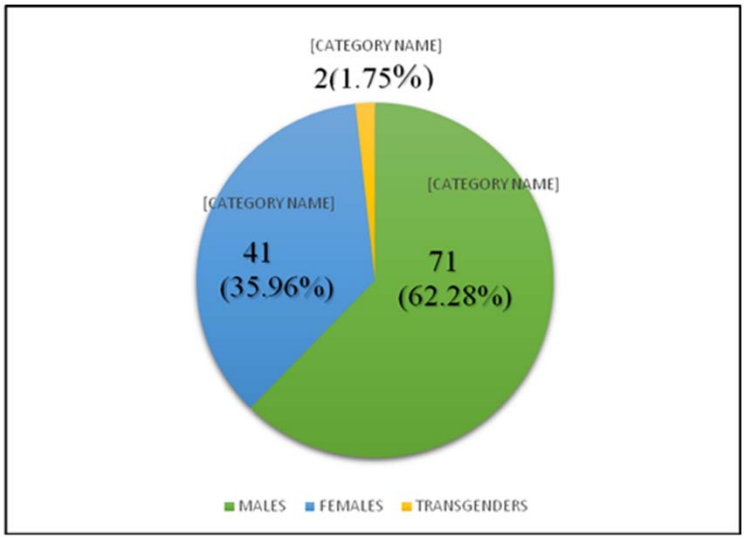

Fig. 1: Gender-wise distribution of study population (n=114)

Table 3: Sex-wise distribution of HIV-HBV and HIV-HCV co-infection (n=114)

	Males	Females	Transgender
Hep B	67 (58.77%)	36 (31.57%)	1 (0.87%)
Hep C	4 (3.50%)	5 (4.38%)	1 (0.87%)
Total	71(62.28%)	41(35.96%)	2(1.75%)

HIV-HBV and HIV-HCV coinfections were predominant in 41-50 y age group. Among them males were 33(28.5%) and females were 18(15.8%). In transgenders it was 31-40 y age group 2(1.7%). Table 4 shows the age-wise distribution of HIV-HBV and HIV-HCV co-infected patients.

Table 4: Age-wise distribution of HIV-HBV and HIV-HCV co-infected patients(n=114)

Age	No. of males	%	No. of female	%	Transgenders	%
0-10	0	0	0	0	0	0
11-20	2	1.75	0	0	0	0
21-30	5	4.38	5	4.38	0	0
31-40	19	16.6	13	11.4	2	1.75
41-50	33	28.5	18	15.8	0	0
51-60	9	7.90	4	3.50	0	0
>61	3	2.63	1	0.88	0	0
Total	71	62.28	41	35.96	2	1.75%

Among 104 HIV-HBV coinfected, viral load at the time of diagnosis is 'below detection level' in 25(24.04%), <250 in 15(14.42%), 251-500 in 9(8.65 %), 501-1000 in 13(12.5 %), 1001-10,000 in 23(22.11 %) and>10, 000 copies/ml in 19(18.26%) of these patients.

In 10 HIV-HCV coinfected cases, the viral load is 'below detection level' in 2(20%), <250 in 1(10%), 251-500 in 1(10 %), 501-1000 in 1(10 %), 1001-10,000 in 4 (40%) and>10,001-100,000 copies/ml in 1(10%). Viral loads at the time of diagnosis of HIV-HBV and HIV-HCV co-infection are shown in the table 5 and fig. 2.

Table 5: Viral load in HIV/HBV and HIV/HCV coinfected patients (n=114) in copies/ml

	TND	<250	251-500	501-1000	1001-10000	>10000	Total
Hep B	25(24.03%)	15(14.9%)	9 (8.65%)	13(12.5%)	23(22.11%)	19(18.26%)	104
Hep C	2(20%)	1(10%)	1(10%)	1(10%)	4(40%)	1(10%)	10
Total	27	16	10	14	27	20	114

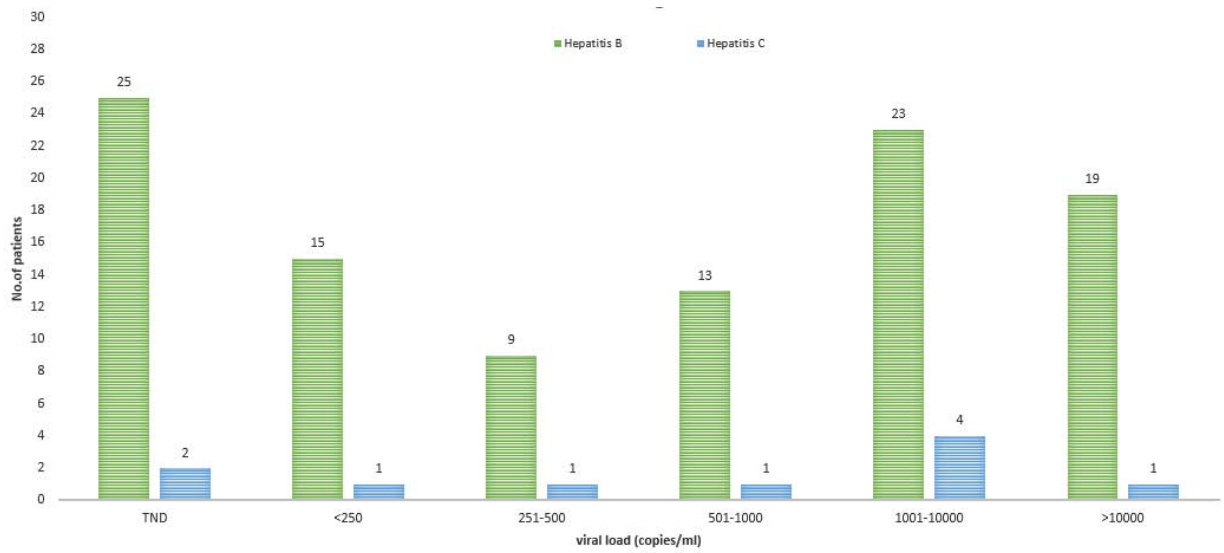

Fig. 2: Bar chart representing the viral load of HIV-HBV and HIV-HCV co-infection

DISCUSSION

HIV continues to be a major public health threat with an additional risk of HBV and/or HCV co-infection [2]. The co-infections are usually underreported in our country [4] and according to many studies, the estimated prevalence of HIV-HBV co-infection rate is 7-30% [6]. Although, there is a high rate of spontaneous clearance of HBV (>90%) in immunocompetent adults, chronic infection develops in 20 percent of adults with HIV infection after exposure to HBV [5].

The prevalence of HCV in HIV-infected persons has ranged between 1.3 and 4.72% [7]. In southern India, prevalence of HCV infection among HIV-infected persons is higher than that in northern India [7]. HIV-HCV co-infection is commoner in patients with low CD4 counts [7].

Out of 5087 samples tested for HIV, 15.6% were found to be seropositive for HIV-HBV and 1.5% for HIV-HCV coinfection. These results are similar to the other studies done by Sonali Bhattar *et al.* 16.7% [3], Saha *et al.* 11.25% [8] for HIV-HBV co-infection and 1.9% for HIV-HCV co-infection by Saha *et al.* [8]. Comparison of HIV-HBV and HIV-HCV co-infection prevalence with other studies is shown in the table 6.

Table 6: Comparison of HIV-HBV and HIV-HCV co-infection prevalence with other studies

Study	No. of HIV positives	Coinfection rate of HIV-HBV	Coinfection rate of HIV-HCV
Shrestha *et al.*	474	14 (2.95%)	86(18.14%)
Saha *et al.*	320	36(11.25%)	6 (1.9%)
Saravanan S *et al.* [9]	500	45(9%)	11(2.2%)
Sarkar *et al.*	1331	78 (5.9%)	-
Desikan and Khan	Meta-analysis	1.89%	
T. Hussain *et al.*	21	2 (9.5%)	1(4.76%)
Sonali Bhattar *et al.*	12	2 (16.7%)	0
Present study	666	104 (15.6 %)	10(1.5%)

The HIV-HBV-HCV co-infections were found to be predominant in males (62.28%). This is clinically significant with a p value of *0.05. This may be because of sexual promiscuity in males and high risk behavior like intravenous drugs use as it poses higher risk of transmission of blood-borne infections [2].

HIV-HBV and HIV-HCV coinfections were predominantly seen in 41-50 y age group in males (28.5%) than females (15.8%). In transgenders, it was 31-40 y age group (1.7%), which shows that the middle-aged population are more affected than younger individuals.

High HBV viral load is a strong predictor of risk of developing hepatocellular carcinoma and plays an important role in treatment success.

Majority of the patients who were infected with HIV-HBV/HIV-HCV was found to have viral loads below detection level at the time of diagnosis. The viral load was below detection level in 24.04% of HIV-HBV and 20% of HIV-HCV coinfected individuals.

CONCLUSION

Screening the high-risk population for these viral infections would aid early detection of co-infections. A timely diagnosis of these infections can minimize downstream adverse health effects, offset rapid disease progression, encourage cure and most importantly, reduce transmission to partners and children. These will cumulatively decelerate co-infection epidemics and will help to decrease the further spread of these blood-borne infections.

Furthermore, monitoring the viral load of HIV at the time of diagnosis or testing for protective levels of HBV/HCV antibodies post-vaccination in uninfected people will help in limiting the course of these diseases and their complications.

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AUTHORS CONTRIBUTIONS

First author of the study Annepu Prasanthi contributed literature search, collected the data, analyzed the data and wrote the first draft of the manuscript. The second author Aruna Bula, contributed conceptual design, data analysis, statistical analysis, literature

survey and corrected the manuscript. The third author Puvvula Kamala, contributed in drafting the manuscript.

CONFLICTS OF INTERESTS

The study declared 'no conflicts of interest'

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