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Prevalence of Infections Caused by Hepatitis B Virus, Hepatitis C Virus and Their Co-Infection in A Tertiary Care Centre, Delhi, India

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ABSTRACT

Background: Hepatitis B and C are the most dominant causes of viral hepatitis, leading to both acute and chronic infections. This study was done to determine the magnitude of the prevalence of Hepatitis B and C and their co-infection.
Subjects and Method: A retrospective cross-sectional observational study was conducted in the Department of Microbiology, North DMC Medical College and Hindu Rao Hospital, Delhi, India from July 2023 to June 2024. All blood samples received from indoor and outdoor patients from all departments were tested by Monolisa HBsAg Ultra and anti-HCV(Ag-Ab Ultra V2) Monolisa by ELISA method. The variables studied were age, sex, in-patient/out-patient, and type of ward. The data was analyzed using the Chi-Square test and Mann-Whitney test.
Results: Of a total of 20,000 blood samples received, 5,358(26.80%) samples were from inpatient department/IPD (2,210 female, 3,148 male) and 14642 (73.20%) and outpatient department/OPD (9,201 female, 5,441 male). 422 samples (2.11%) were positive for HBsAg (210 IPD -77 female, 133 male; 212 OPD - 121 female, 91 male) and 614 (3.07%) for anti-HCV (408 IPD - 135 female, 273 male; 206 OPD - 100 female, 106 male). Co-infection of Hepatitis B and C viruses was seen in 40 patients, 31 IPD (10 female, 21 male) and 9 OPD (1 female, 8 male). Maximum cases were seen from medicine wards among IPD patients of HBsAg positive, anti-HCV positive, and co-infection and in the age group 41-60 years in all three categories.
Conclusion: Screening for HBV and HCV infection is compulsory so that preventive measures are implemented to improve public health.

Keywords: Hepatitis B virus (HBV), Hepatitis C virus (HCV), HBsAg, Anti-HCV Ab

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BACKGROUND

Viral hepatitis has emerged as a major public health problem after tuberculosis and HIV (Parveen et al., 2020). It is caused by Hepatitis A, B, C, D, and E viruses. Hepatitis A and E, transmitted through the faecal-oral route, usually present as acute infections which may occur sporadically or as out-breaks. Hepatitis B and C, transmitted predominantly by percutaneous or mucosal exposure to infected blood and various body fluids, can lead to chronic infection and progress to cirrhosis and hepatocellular carci-

noma (National Program for Surveillance of Viral Hepatitis, 2021).

Hepatitis C and B were shown to be the most dominant causes of hepatitis in several groups, including the public, children, healthcare workers, and individuals with acute and chronic liver disorders (Maqsood et al.,2023). HBV and HCV infections are similar in distribution and both lead to liver cirrhosis and hepatocellular carcinoma and are difficult to cure. Hepatitis B virus is a DNA virus and it causes both acute and chronic infections. Acute infection lasts for less than 6 months and is associated with acute inflammation and hepatocellular necrosis. Chronic infection is the persistence of Hepatitis B surface Antigen (HBsAg) in blood or serum for more than 6 months. Chronicity is mostly observed in neonates and younger children (Maqsood et al.,2023).

Hepatitis C virus is a single-stranded RNA virus. Acute infection is associated with milder symptoms and in chronic cases, 5-20 % of patients develop cirrhosis and 25% of them develop end-stage liver disease and hepatocellular carcinoma (Parveen et al., 2020). According to WHO, globally 350 million and 71 million people are living with HBV and HCV infection respectively (Bhadoria et al.,2021). In 2015, 1.34 million fatalities worldwide were caused by viral hepatitis, and the majority of deaths were caused by chronic liver disease or primary liver cancer (Kumar et al., 2023).

The National Program for Surveillance of Viral Hepatitis (NPSVH) (2021) at the National Centre for Disease Control (NCDC), DteGHS, MoHFW aims to obtain nationally representative data to assess the disease burden in India for Hepatitis B and Hepatitis C to assist in optimal planning for prevention and management of viral hepatitis in the country. According to this, the national seroprevalence of Hepatitis B was 0.95% (0.89-1.01) among women aged 15-49 years and men aged 15-54 years in 2015-16. In India, in 2015 approximately 40 million people were chronically infected with Hepatitis B and 6-12 million people with Hepatitis C (Parveen et al., 2020).

The WHO Global Hepatitis Strategy (2023) is supported by all WHO member states and they aim to reduce new hepatitis infections by 90% and deaths by 65% between 2016 and 2030. Although HBV/ HCV co-infection is not uncommon, its epidemiology is poorly defined. Co-infection occurs when two or more reproducing organisms coexist in the same host. HBV and HCV have significantly different life cycles, even though they like to multiply in hepatocytes. HCV replicates in the cytoplasm of hepatocytes, while HBV replicates in the nucleus (Liu and Kao, 2020). They may interact in co-infected cells and elicit unique viral expression and serologic patterns because they both include RNA replicate intermediates (Asim et al., 2016).

As compared to mono-infected patients, HBV and HCV co-infected persons tend to have more severe liver injury, a higher probability of liver cirrhosis and hepatic decompensation, and a higher incidence of hepatocellular carcinoma (Grewal et al., 2018). Co-infection is usually seen in highly endemic areas or among those at great risk of infection, like those who inject drugs or homosexual men (Maqsood et al.,2023). Patients who have hepatitis C and B need vigilant monitoring and potent antiviral drugs. Also, WHO is urging countries to take quick action to increase knowledge, diagnosis/ testing, and treatment services for hepatitis. So, there is a need to study the magnitude of the disease in order to clearly understand its prevalence; thus, this study was undertaken. To find the prevalence of infections caused by Hepatitis B virus, Hepatitis C virus, and their co-infection in a tertiary care centre in Delhi, India.

SUBJECTS AND METHOD

1. Study Design

A retrospective cross-sectional observational study was conducted in the Department of Microbiology, North DMC Medical College & Hindu Rao Hospital, Delhi, India over 1 year from July 2023 to June 2024.

2. Population and Sample

All the indoor and outdoor patients' blood samples sent from various departments were included. Hemolysed samples, duplicate samples, and occult HBV infection were excluded.

3. Study Variables

The variables studied were age, sex, in-patient or out-patient, and type of ward.

4. Operational Definition of Variables

The details were taken from the blood test requisition forms.

Age: The age of the patients examined in this study.

Sex: The characteristic of the patient categorized as male or female

In-patient/ out-patient: in-patient, referring to those admitted to the hospital for at least 24 hours under medical supervision, or out-patient, referring to those who visit the healthcare facility for examination or treatment without requiring hospitalization.

Type of ward: The category of hospital unit or department where an in-patient receives treatment.

5. Study Instrument

Around 5 ml of the patient's blood was collected. From it, the serum was separated out and was divided into two aliquots for performing Monolisa HBsAg Ultra and anti-HCV (Ag-Ab Ultra V2) Monolisa by ELISA method.

6. Data Analysis

Statistical analysis was done using the Chi-Square test and the Mann-Whitney test.

7. Research Ethics

Permission was taken from the Scientific Committee.

RESULTS

In total, 20,000 blood samples were received in the Department of Microbiology from outpatient departments (OPD) and in-patient (IPD) from various departments during a period of one year from July 2023 to June 2024. Out of the 20,000 samples received, 5,358 (26.80%) samples were from IPD and 14,642 (73.20%) from OPD. Amongst these 20,000 samples received, 11,411 (57.06%) were from female and 8,589 (42.94%) from male patients. Of these 5358 IPD samples, 2,210 (41.25%) were from females and 3,148 (58.75%) were from male patients. Out of 14,642 OPD samples, 9,201 (62.84%) samples were from female patients and 5,441 (37.16%) were from male patients (Table 1).

Table 1. Results of Total Blood Samples Received

Samples received	Female Patients Tested		Male Patients Tested		Total
	n	%	n	%	
In-patient	2,210	19.37	3,148	36.65	5,358
Out-patient	9,201	80.63	5,441	63.35	14,642
Total	11,411		8,589		20,000

Amongst 20000 patients' samples tested for HBsAg and anti-HCV, 422 (2.11%) were positive for HBsAg, and 614 (3.07%) were positive for anti-HCV. Out of 5358 IPD samples tested for HBsAg, 77 specimens from female patients were positive, and 133 specimens from male patients. Of 14,642 samples from the OPD patients tested for HBsAg, 121 specimens from female patients were positive, and 91 from the male patients. In total, 422 patients' samples were positive for HBsAg from the 20,000 samples tested;

out of these 210 (49.76%) specimens from IPD patients were positive, and 212 (50.24%) specimens were positive from OPD patients (Table 2 and Table 3).

Table 2. Results of Samples Tested for HBsAg

HBsAg	In-patient				Out-patient				Total
	Female		Male		Female		Male		
	n	%	n	%	n	%	n	%	
Positive	77	3.48	133	4.22	121	1.32	91	1.67	422
Negative	2,133	96.52	3,015	95.78	9,080	98.68	5,350	98.33	19,578
Total	2,210	100	3,148	100	9,201	100	5,441	100	20,000

Table 3. Results of Sample Tested for HBsAg

HBsAg	Positive		Negative		Total
	n	%	n	%	
In-patient	210	3.92	5,148	96.08	5,358
Out-patient	212	1.45	14,430	98.55	14,642
Total	422	2.11	19,578	97.89	20,000

Out of 5,358 IPD patients' samples tested for anti-HCV, 273 specimens from male patients were positive, and 135 specimens from female patients. Of 14,642 samples from the OPD patients tested for anti-HCV, specimens were positive for 100 female patients and 106 male patients. In total, 614 patients were positive for anti-HCV from the 20,000 samples tested; out of these, IPD specimens were positive for 408 (66.45 %) patients and 206 (33.55%) specimens were positive from OPD patients (Table 4 and Table 5).

Table 4. Results of Samples Tested for Anti-HCV

Anti-HCV	In-patient				Out-patient				Total
	Female		Male		Female		Male		
	n	%	n	%	n	%	n	%	
Positive	135	6.11	273	8.67	100	1.09	106	1.95	614
Negative	2,075	93.89	2,875	91.33	9,101	98.91	5,335	98.05	19,386
Total	2,210	100	3,148	100	9,201	100	5,441	100	20,000

Table 5. Results of Samples Tested for Anti-HCV

HBsAg	Positive		Negative		Total
	n	%	n	%	
In-patient	408	7.61	4,950	92.39	5,358
Out-patient	206	1.41	14,436	98.59	14,642
Total	614	3.07	19,386	96.93	20,000

Amongst these 20,000 samples, co-infection of hepatitis C and B viruses was seen in 40 patients out of which 31 were from IPD and 9 from OPD. Co-infection was more common among the male patients in both IPD and OPD. In IPD 21 male patients (67.74%) had co-infection while female patients were 10 (32.26%); among OPD 8 male (88.89%) and 1 female (11.11%) (Table 6).

Table 6. Results of Co-infection with HBsAg and Anti-HCV

HBsAg + Anti HCV Positive	Female		Male		Total
	n	%	n	%	
In-patient	10	90.91	21	72.41	31
Out-patient	1	9.09	8	27.59	9
Total	11	27.5	29	72.5	40

Among the total positive HBsAg cases in IPD patients, the maximum positive cases were from medicine wards (90; 42.86 %) followed by ICU (35; 16.67%), surgery wards (25; 11.90%) and CCU (17; 8.10%). Among the total positive anti-HCV cases in IPD patients, the maximum positive cases were from medicine wards (119; 29.17 %) followed by thalassemia ward (100; 24.51%), ICU (44; 10.78%), surgery wards (34; 8.33%) and dialysis (27; 6.62%). Among the positive co-infection (HBV & HCV viral infections) cases, the maximum cases were from the medicine ward (11) followed by dialysis (7), CCU (5), and ICU (3) (Table 7).

Table 7. Ward-wise Distribution of Positive Cases

Department	HbsAg positive				Anti-HCV positive				HbsAg+Anti-HCV positive			
	Female		Male		Female		Male		Female		Male	
	n	%	n	%	n	%	n	%	n	%	n	%
Medicine	30	41.0	60	43.7	33	24.4	86	31.5	5	41.6	6	31.5
Surgery	14	19.1	11	8.0	15	11.1	19	6.96	0	0	1	5.2
Orthopaedics	1	1.3	15	10.9	8	5.9	26	9.52	0	0	2	10.5
Obs & Gynae	0	0	5	3.6	6	4.4	0	0	0	0	0	0
Pediatrics	3	4.1	4	2.9	4	2.9	8	2.93	1	8.3	0	0
Eye	1	1.3	1	0.7	2	1.4	2	0.73	0	0	0	0
Thalassemia	2	2.7	2	1.4	29	21.4	71	26.01	1	8.3	0	0
Dialysis	2	2.7	7	5.1	10	7.4	17	6.23	1	8.3	6	31.5
ICU	12	16.4	23	16.7	16	11.8	28	10.26	2	16.6	1	5.2
CCU	8	10.9	9	6.5	10	7.4	16	5.86	2	16.6	3	15.7
NICU	0	0	0	0	2	1.4	0	0	0	0	0	0
Total positive	73	34.7	137	65.2	135	33.0	273	66.9	12	38.7	19	61.2

Among the positive HBsAg cases, maximum cases were between 41-60 years of age (229; 54.26%), followed by 13-40 years of age (99; 23.46%), then >60 years of age group (84; 19.90%), and then 0-12 years of age groups (10; 2.37%). Among the age group of 41-60 years of age, 148 (35.07%) males were positive and 81 (19.19%) females were positive. In the age group of 13-40 years, 73 females (17.30%) were positive and 26 males (6.16%) were positive. Among the positive anti-HCV cases, maximum cases were between 41-60 years of age (217; 35.34%), followed by >60 years of age (186; 30.29%), then 13-40 years of age group (180; 29.31%), and then 0-12 years of age group (31; 5.05%). In the 41-60 years of age group, 140 (22.80%) males and 77 (12.54 %) females were positive respectively. In the age group of >60 years, 115 males (18.73%) and 71 females (11.56%) were positive. Among the co-infection cases, maximum positive cases were from 41-60 years of age group (21; 52.50%) followed by >60 years of age group (15; 37.50%) (Table 8).

Table 8. Age-wise Distribution of Positive Cases

Age (years)	HbsAg positive					Anti-HCV positive					HbsAg+Anti-HCV positive				
	Female		Male		Total	Female		Male		Total	Female		Male		Total
	n	%	n	%		n	%	n	%		n	%	n	%	
0-12	2	1.0	8	3.5	10	8	3.4	23	6.07	31	0	0	1	3.4	1
13-40	73	36.8	26	11.6	99	79	33.6	101	26.6	180	0	0	3	10.3	3
41-60	81	40.9	148	66.0	229	77	32.7	140	36.9	217	3	27.2	18	62.0	21
>60	42	21.2	42	18.7	84	71	30.2	115	30.3	186	8	72.7	7	24.1	15
Total positive	198	46.9	224	53.0	422	235	38.2	379	61.7	614	11	27.5	29	72.5	40

DISCUSSION

Though reports on the prevalence of HBV and/or HCV infection among blood donors (Arora et al., 2010) and in patients with certain clinical conditions (Ali et al., 2008; Mysorekar et al., 2008) are available, prevalence among the general population in India is sparse. Out of 20,000 patient samples tested in our study, in-patient specimens received were more in males (3,148; 36.65%) than in females (2,210; 19.37%), which is statistically significant (p<0.001). Among the OPD patients, specimens received were more in females (9,201; 80.63%) than males (5441; 63.35%), which is also statistically significant (p<0.001). Among the HBsAg positive cases, the mean age group of the patients was 49 years while among the positive anti-HCV cases mean age group of the patients was 48 this is similar to the study done by Grewal et al. (2018) (with a mean age of 47.5 years), but is not similar to the study done by Sharma et al. (2017) (mean age was found to be 55 years). Among the positive cases in our study, 614 (3.07%) were positive for anti-HCV which is statistically significant in IPD cases as compared to OPD cases (p <0.00001); and 422 (2.11%) were positive for HBsAg which is statistically significant in OPD as compared to IPD cases (p<0.001).

Prevalence of hepatitis B has been reported to range between 0.1%-11.7% (average is between 2-8%) in most of the studies, placing India in the intermediate range of hepatitis B endemicity; which is equal to our study prevalence range results. The prevalence of hepatitis B in our study was found to be 2.11%, which is similar to that of a study done by Pilli et al. (2020) which showed a prevalence of 2%. Suzuki et al. (1994) reported a prevalence rate of 4%, while Uppal et al. (2009) reported the prevalence of HBV in an urban slump of northern India to be 10.38% and a study done by Sharma et al. (2017) showed the prevalence of HBV as 9%; which is on the higher side as compared to our study.

The prevalence of hepatitis C in our study was 3.07%, which is similar to the study done by Pilli et al. (2020) which showed a prevalence of 3.1%. But it is on the higher side as compared to studies done by Chowdhury et al. (2003) who showed a prevalence of 0.87% and Bhardwaj et al (2014) who showed a prevalence of 0.68% in Tamil Nadu. However, the studies done by Sharma et al. (2017), Sood et al. (2012) and Parveen et al. (2020) showed a prevalence of 5.5%, 5.2%, and 5.23% respectively, which are on the higher side as compared to our study.

Among the positive co-infection cases in our study, prevalence was found to be 0.20%; which is similar to the study done by Pilli et al. (2020). Among the positive cases in our study, co-infection was more in males (72.5%) as compared to females (27.5%) which is statistically significant (<0.001), and is similar to the study done by Pilli et al. (2020) and also statistically significant among the IPD cases as compared to OPD cases (p <0.001). Among ward-wise distribution of HBsAg, Anti-HCV, and co-infection cases, maximum cases were positive from the medicine ward; which is statistically significant (p<0.001). Among the age-wise distribution of positive cases, maximum cases were in the age group of 41-60 years of age in HBsAg positive, anti-HCV positive, and co-infected cases and statistically significant (p <0.001). Grewal et al. (2018) showed a majority of the HBsAg-positive cases were in the age group of 21-40 years, while maximum anti-HCV cases belonged to the age group of 41-60 years and this is similar to our study. But this is not similar to the study done by Pilli et al. (2020) who showed HBV in the majority of patients in the 20-30 years age group;

whereas HCV and co-infection were more commonly seen in the 61-70 years age group.

The only limitation of our study was that we did not identify risk factors associated with both HBV and HCV infections. Screening of high-risk individuals is compulsory because of a comparatively higher proportion of HCV and HBV infection among hospital-based studies and also because it is a major public health concern in the rural population. The risk of liver failure increases with co-infection of both HCV and HBV. Knowing the prevalence of HBV and HCV infection can help in applying preventive measures like safe injections, safe blood supply, health education about awareness of safe sex, early detection of infection, and implementation of HBV vaccination in high-risk groups leading to improved public health.

AUTHOR CONTRIBUTION

Manoj Kumar has collected and analysed all the data and written the manuscript. Sanjay Jain has critically reviewed the manuscript.

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None.

CONFLICT OF INTEREST

None.

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