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Seroprevalence of Hepatitis B and C virus Infection Among Clinically Suspected Patients At State Sector Hospitals In Sri Lanka; A Laboratory Based Retrospective Study

A.I.K.Mahanama[1], B.Samaraweera[2], T.T.Iroshanie[3], S.S.S.A.K.Seethagala[4],*J.I.Abeynayake[5]
[1,2,3,4,5] Medical Research Institute, Colombo 08, Sri Lanka
Correspondent Author: janakiiabeynayake@yahoo.com
Email address of other authors: adhyanaindunil@gmail.com,buddiyasasi@gmail.com

Abstract:

The public health threat posed by viral hepatitis globally is being planned to be eliminated by the year 2030 and, Hepatitis B virus (HBV) and Hepatitis C virus (HCV) infections accounts for more than 90% of the worldwide viral hepatitis burden.
Sri-Lankans has an increased risk of acquiring HBV and HCV infections through travel and trade as many neighboring countries report intermediate to high HBV and HCV prevalence. Data regarding the prevalence of these infections, which is essential for planning of country specific elimination strategies are scarce and is in high demand. Hence, the current study aims at describing the prevalence and associated factors for HBV and HCV infections in patients tested at Medical Research Institute (MRI), the premier institution for diagnosis of viral hepatitis in Sri-Lanka which receives samples from all around the country.
A retrospective study was conducted using all samples received and tested at MRI for HBsAg and HCV antibody from 2016 to 2018, using a commercially validated HBsAg and anti-HCV antibody ELISA kits respectively. Clinical and epidemiological data were collected using the accompanying request form.
A total of 36,007 patient samples were tested for HBsAg and 15,280 for anti HCV antibodies.
Overall HBsAg seroprevalence was 1.56% for the study period. Majority (65.06%) of the positives were males and majority (23.58%) was in 31-40 years age category. Highest numbers of positives (53%) were reported from the Colombo district. Most (23.8%) were chronic careers, 15.8% had acute hepatitis and 11.8% had chronic liver disease due to HBV infection. Out of all positives 5.1% were pregnant at the time of testing.
Anti HCV seroprevalence for the study period was 1.81%. Males were predominant (65.9%) with mean age being 39 years (SD 16.39). Seropositive cohort consisted of multiple risk groups including patients with multiple blood transfusions, IV drug use, multiple sexual partners, and haemodialysis.
Majority of positive samples were received from Colombo, which represented 54.5% of the total.
The study results confirmed the HBV and HCV infection burden in Sri Lanka and the need to strengthen and expand preventive and control measures in order to achieve the WHO declared elimination goals by the year 2030.

Keywords

Hepatitis B, Hepatitis C, Prevalence, HBsAg, anti HCV antibodies, Sri Lanka,

1. Introduction

Viral hepatitis has caused 1.34 million deaths worldwide in the year 2015 and Hepatitis C virus (HCV) and hepatitis B virus (HBV) accounts for more than 90% of this burden due to chronicity and associated complications of the infection [1,2]. Identifying the gravity of the situation the Global Health Sector Strategy (GHSS) on viral hepatitis 2016-2021 initiated a campaign to eliminate the public health threat by viral hepatitis (90% reduction of new infections and 65% reduction of mortality as compared to the rates in 2015 as baseline) by the year 2030 [1].

HBV is a hepatotrophic virus transmitted by exposure (percutaneous/mucosal/sexual/transfusion) to infected blood and/or body fluids and also from mother to child. More than 95% of adults who get the infection clear it off, however perinatal acquisition may lead to chronicity in around 80-90% of the exposed [3]. HBsAg is the first serological marker of infection and persists until the infection is cleared off and chronic infection is defined by the persistence of HBsAg for more than six months in serum. Chronic HBV infection may lead to progressive liver disease, cirrhosis and hepatocellular carcinoma. Anti-viral therapy does not cure HBV infection, but slows the progression /incidence of complications. Hepatitis B vaccine has been recommended by World Health Organization (WHO) as the main mode of prevention of HBV infection and is indicated to all infants, high risk categories for infection as well as to everyone living in low-intermediate endemic countries [3].

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It is estimated that 257 million people are HBsAg positive globally and countries are divided in to three categories depending on their HBsAg prevalence; high prevalence (>8%), intermediate prevalence (2-8%) and low prevalence (<2% [6]. Highest HBV prevalence is reported in Western Pacific and African WHO regions while the lowest is from Americas and European region. According to the WHO data the South East Asia region, to which Sri-Lanka belongs to reports high-intermediate prevalence [3].

HCV is also a hepatotrophic virus mainly transmitted by percutaneous exposure to infected blood or body fluids and the chronicity of infection is around 60-80 % [4]. Chronic HCV infection may lead to chronic liver cell disease (CLCD), cirrhosis of liver, hepatocellular carcinoma, liver failure and also death [5].The serological marker of HCV infection is detection of HCV antibodies and once positive it remains positive for life whether cured or not. It does not indicate immunity [4]. Confirmation of HCV infection is mainly by demonstrating the presence of HCV specific RNA in blood. .Majority (>95%) of cases with HCV infection can be cured by early initiation of antiviral therapy [4].

The global burden of HCV infection is around 71 million and there had been 23.7 new HCV cases/100000 people globally [4]. The world is being divided in to three categories based on HCV prevalence; <1.5%-low prevalence, 1.5-3.5% - moderate prevalence and > 3.5% high prevalence [7]. HCV affects all WHO regions with most affected regions being WHO Eastern Mediterranean and Europe regions [2, 4].

HBsAg prevalence in Sri-Lanka ranges from 0.1 to 2.5% in various high risk communities for HBV infection [8] and this rate has placed Sri-Lanka in low to intermediate prevalence level [10]. However most of the neighboring countries report intermediate-high HBV prevalence thus Sri-Lanka is at high risk of increasing the HBV prevalence rate over years due to travel, trade and tourism. Many strategies are undertaken in par with global elimination effort to control the infection which includes immunization, national policies on injection safety in health-care setting and transfused blood/blood products safety.

Seroprevalence of HCV infection in Sri Lanka was 2.1% among patients with alcoholic cirrhosis [9], 6.9% among prison inmates [10], 1.06% among healthy blood donors [11) and 5% in patients with multiple blood transfusions [12). However these studies were conducted using a small sample size and results from larger scale studies are lacking.

Epidemiology of HBV and HCV infections are ever changing due to globalization which facilitate large-scale spread of the infection and also due to various preventive measures that are being implement to combat the spread [13). Therefore periodical re-evaluation of prevalence rate is important in the context of planning for elimination of viral hepatitis to plan for effective health strategies. At the same time it is prudent to have seroprevalence data of HCV and HBV infections from larger scale studies analyzing a large number of samples which represent the whole country. Medical research institute is the premier laboratory testing for viral hepatitis in the country and receives a large number of samples from all around the country. Thus this study was conducted aiming to determine the seroprevalence of HBV and HCV infections in clinically suspected samples tested at MRI over a period of three years and to characterize the socio-demographic details, clinical presentations and distribution in to identified risk groups for infection among the HBV and HCV positive samples.

2. Materials and Methods

A retrospective analysis was conducted using the results of all samples (serum/plasma) received for testing at MRI for HBsAg and HCV antibody from 2016 to 2018. Testing for HBsAg and HCV antibodies were carried out using commercially validated HBsAg and HCV ab Enzyme Immuno Assays (EIA). HBsAg testing was carried out mostly by the kit HBsAg ULTRA with a sensitivity of 100% and specificity of more than 99% and the HCV antibody testing was by Monolisa Anti HCV PLUS kit with a sensitivity of 100% and specificity of more than 99%. Validation, analysis and interpretation of results were as per the manufacturer's instructions. HBsAg and HCV antibody positive samples were repeated in duplicate and a second sample was tested for if and when necessary. Testing algorithm for HBsAg and HCV antibodies are outlined by figure 1. A sample was entered as positive only after confirmation following repeat testing. HCV antibody positive samples were reflexed tested for HCV RNA.

However, HCV RNA detection results were not included in the study as the study aims to determine HCV antibody prevalence. Scio-demographic details, data on clinical presentation and inclusion in to risk groups for infection of positive samples were gathered using the accompanying request forms. For HBsAg positive patient's data were available for all three years, however for HCV antibody positive patients' data collection was done for the positive samples in the year 2017 and 2018 only as request forms for preceding years were unavailable. Furthermore, due to incompleteness of request forms

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data for all the observed variables of all positive patients was not able to gather, thus analysis was carried out using the number of patients included for each variable and this would be shown in the results section. Statistical analysis was by SPSS most recent version and descriptive statistics was used to present the data.

3. Results

3.1. HBsAg and HCV sero-prevalence

A total of 36,007 patients were tested for HBsAg during the study period. Overall HBsAg seroprevalence was 1.56% for the study period. It showed a fluctuating trend with 2.2% in 2016, 1.32% in 2017 and 1.39% in 2018.

For HCV antibodies a total of 15,280 patients were tested during the study period. In 2016 HCV antibody prevalence was 1.7%, 2017 - 1.98%, 2018 – 1.75% with the overall prevalence being 1.81%.

Table 1. HBsAg and HCV antibody prevalence from 2016 to 2018

Year	HBsAg			HCV antibody		
	Total	Positive	%	Total	Positive	%
2016	8707	192	2.20%	2337	40	1.7%
2017	12044	160	1.32%	4239	84	1.98%
2018	15256	213	1.39%	8704	153	1.75%
Total	36,007	565	1.56%	15,280	278	1.81%

3.2. Socio-demographic characteristics of HBsAg positive patients

Socio-demographic details are depicted in table 2. Majority (65.06%) of HBsAg positives were males. Age ranged from 1 month to 85 years and mean age was 41.34 (SD 16.4) years. Majority (23.58%) were in 31-40 years age category. Highest numbers of positives (53%) were reported from the Colombo district while Galle reporting 8.7% which is the second highest in the country. Cases were not reported from Kegalle, Kilinochchi, Mannar and Matale.

3.3. Clinical presentation of HBsAg positives

Majority (23.8%) of the positives were of chronic HBV careers without any evidence of liver damage, 15.8% had acute hepatitis and 11.8% had chronic liver disease (CLD) due to HBV infection. Out of all positives 5.1% were pregnant at the time of testing and were identified as part of ante natal HBV screening program. The categorization of the positives based on their clinical presentation is expressed in Table 2.

3.4. Distribution of HBsAg positives in to risk groups for acquisition of infection

Categorization of high risk groups and distribution of HBsAg positives is summarized in Table 2 and 28.9% of them were patients needing multiple transfusions.

Table 2. Socio-demographics, clinical presentation and distribution in to risk groups of HBsAg positive patients

Variable	Result
Mean age(SD) (Range) n=458	41.34 years (16.4) (1 month -85 years)
Age break down (n=458)	
< 1	05 (1.09%)
1-10	08 (1.74%)
11-20	33 (7.2%)
21-30	85 (18.55%)
31-40	108 (23.58%)
41-50	77 (16.81%)
51-60	76 (16.59%)
61-70	51 (11.13%)
71-80	14 (3.05%)
81-90	01 (0.26%)
Gender (n=395)	
Male	257 (65.06%)
Female	138 (34.94%)
Distribution of cases as per district (n=366)	
Ampara	5 (1.4%)
Anuradhapura	27 (7.4%)
Badulla	7 (1.9%)
Batticaloa	9 (2.4%)
Colombo	194 (53%)
Galle	32 (8.7%)
Gampaha	16 (4.4%)
Hambantota	7 (1.9%)
Jaffna	14 (3.8%)
Kalutara	6 (1.6%)
Kandy	6 (1.6%)
Kegalle	0
Kilinochchi	0
Kurunegala	20 (5.5%)
Mannar	0
Matale	0
Monaragala	4 (1.1%)
Mullativu	1 (0.3%)
Nuwara Eliya	1 (0.3%)
Polonnaruwa	2 (0.5%)
Puttalam	5 (1.4%)
Ratnapura	4 (1.1%)
Trincomalee	3 (0.8%)
Vavuniya	1 (0.3%)
Matara	2 (0.5%)
Clinical presentation (n=298)	
Acute hepatitis	47 (15.8%)
Antenatal screening	16 (5.1%)
Blood donor screening	36 (12.1%)
Carcinomas	21 (7.1%)
Chronic HBV carriers	71 (23.8%)
Chronic liver cell disease	35 (11.8%)

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Contact screening	9 (3%)
Medical screening	15 (5%)
Chronic Kidney Disease	5 (1.8%)
IV drug addicts	2 (1%)
Source; occupational exposure	7 (2.3%)
Multiple transfusions	10 (3.3%)
Patients with STI	3 (1.1%)
Screening before surgical procedures	10 (3.9%)
Treatment monitoring	11 (3.8%)
Distribution of HBsAg positives based on risk groups for infection (n=38)	
Dialysis patients	5 (13.1%)
Household/sexual contacts of HBV infected	9 (23.7%)
IV drug users	2 (5.4%)
Individuals having multiple sexual partners	3 (7.9%)
People requiring multiple transfusions	11 (28.9%)
Individuals at occupational risk	4 (10.5%)
Inmates of custodial institutions	4 (10.5%)

IV-Intravenous, STI-Sexually Transmitted Infections

3.5. Socio-demographic characteristics of HCV serology positive patients

Analysis of socio-demographic characteristics was carried out on the positive samples received in the year 2017 and 2018 and the details are expressed in Table 3. Males were predominant (65.9%) among the HCV seropositives. Mean age was 34 years (SD 16.26) with the age ranging from 2 to 70 years and most (27.9%) of the HCV seropositives were in 21-30 years of age range. Majority of the positive samples were received from Colombo, which represented 54.5% of the total. Kurunegala and Gampaha had the second highest (11.6%) positive rate with Anuradhapura being the third (10.3%).

3.6. Clinical presentation of HCV antibody positive patients

Among the positive group, majority (56.16%) of positives was in patients with multiple blood transfusions and, 15.75% were patients with a history of STI while 10.28% were with chronic liver cell disease.

3.7. Distribution of HCV antibody positives based on risk groups for HCV infection

The seropositive cohort had individuals belonging to multiple HCV risk groups and most (75.22%) were individuals needing multiple blood transfusions. There were IV drug users (1.84%) and individuals with a history of HIV or other sexually transmitted infections (21.1%) as well.

Table 3: Socio-demographic characteristics, clinical presentations and distribution of HCV positives based on high risk groups for HCV infection

Variable	Result
Mean age (SD) (Range) n=208	34 years (16.26) (2 -70 years)
Age break down (years) n=208	
< 10	8 (3.8%)
11-20	41 (19.7%)
21-30	58 (27.9%)
31-40	32 (15.4%)
41-50	40 (19.2%)
51-60	13 (6.3%)
61-70	16 (7.7%)
Gender (n=179)	
Male	118 (65.9%)
Female	61 (34.1%)
Distribution of cases as per district (n=224)	
Ampara	01 (0.4%)
Anuradhapura	23 (10.3%)
Badulla	01 (0.4%)
Batticaloa	00
Colombo	122 (54.5%)
Galle	04 (3.2%)
Gampaha	26 (11.6%)
Hambantota	00
Jaffna	07 (3.2%)
Kalutara	01 (0.4%)
Kandy	06 (2.7%)
Kegalle	01 (0.4%)
Kilinochchi	00
Kurunegala	26 (11.6%)
Mannar	00
Matale	00
Monaragala	00
Mullativu	00
Nuwara Eliya	00
Polonnaruwa	00
Puttalam	00
Ratnapura	03 (1.3%)
Trincomalee	00
Vavuniya	00
Clinical presentation (n=146)	
Acute hepatitis	8 (5.48%)
Chronic liver cell disease	15 (10.28%)
Medical screening	7 (4.8%)
Chronic Kidney Disease	5 (3.42%)

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IV drug addicts	3 (2.05%)
Multiple transfusions	82 (56.16)
Patients with a history of STI	23 (15.75%
Screening before surgical procedures	2 (1.38%)
PUO	1 (0.68%)
Distribution of HCV antibody positives based on risk groups for acquisition of infection (n=109)	
IV/intranasal drug users	02(1.84%)
Recipients of multiple transfusions	82 (75.22%)
Sexual partners of HCV infected	0
People with HIV/other STIs	23 (21.1%)
Children born to HCV positive mothers	0
Prisoners/previously incarcerated persons	2 (1.84%)
People with tattoos/piercings	0

IV-Intravenous, PUO-Pyrexia of Unknown Origin, STI-Sexual Transmitted Infections

4. Discussion

The current study was carried out with the goal of determining the sero-prevalence of HBV and HCV infections in clinically suspected samples tested at Medical Research Institute, the premier institution for testing of viral hepatitis in the country.

More than 10,000 samples per infection were tested during a period of three years and an overall HBsAg prevalence of 1.56% and HCV sero-prevalence of 1.81% was determined.

The prevalence of HBsAg determined by the study is less than the global estimated prevalence of 3.61 % and is comparable to that in the South East Asian Region which is 1.9% [14]. However when comparing with countries in the region with which Sri-Lanka has close trade and tourism relationships the rate is very much low. Zampino et al in 2015 reports in their review that India has a HBV sero-prevalence of 3.7%, china 7.18%, Singapore 3.6% and South Korea 4%. At the same time the rate in the study is comparable to previous studies conducted in high risk groups in Sri Lanka [8]. The determined sero-prevalence places the country in low prevalent category with increased risk of acquisition of the infection from neighboring countries [15]. HBsAg prevalence shows a fluctuating trend over the three years with a prevalence of > 2% (intermediate prevalence category) reported in the year 2016. However, by the year 2017 and 2018 the rate declines to 1.32% and 1.39% respectively which may be attributable to island wide activities carried out to reduce HBV transmission among high risk groups which includes HBV immunization campaigns, promoting safe sexual practices and safe injection usage among IV drug addicts, strengthening of infection control practices and injection safety in health care settings and ensuring transfused blood and blood product safety by pre transfusion vigorous screening practices.

HCV sero-prevalence reported in the study places Sri Lanka in the moderate prevalence category [7]. It is slightly less the global prevalence rate of 2.5% and regional rate of 2.8% [16]. However this rate is higher than that for the general population in India (0.8%) while being very much lesser than the rate in Pakistan (6.7%). The difference between India and Sri-Lanka may be attributable to the differences in study populations which is the general population in India in contrast to the high risk populations in Sri Lanka. When comparing with similar studies conducted in Sri-Lanka the rate determined in the current study is low [9, 10, 12]. In contrast to HBsAg prevalence HCV sero-prevalence depicts a constant trend over the three years with the lowest prevalence reported in 2016.

Analysis of socio demographic data depicts similar gender distribution for HBV and HCV infection. Majority of the affected were adults (mean age for HBsAg - 41 years, mean age for HCV antibody-34 years) expressing that in Sri-Lanka these infections are still confined to adult population, thus exerting a negative impact on the work force in the country. The involvement of the pediatric population which is indicative of poor elimination outcomes is less presently. However, five of the HBsAg positives were less than 1 year of age and 5.1% of the positives were pregnant at the time of testing (antenatal screening), indicating the presence and possibility of perinatal transmission respectively, which has a 80-90% risk of developing in to chronic infection and emphasizing the need for implementing routine ante-natal screening of all pregnant mothers to eliminate mother to child transmission of the infection.

Colombo district being the commercial capital of Sri-Lanka reports the highest district wise sero-prevalence rates for both infections. The second in the list for HBsAg is Galle and for HCV it is Kurunegala and Gampaha. In the country, Kurunegala district harbors a large population of thalassemia patients who are requiring multiple transfusions thus placing them at increased risk of acquiring the infections.

High risk groups for HBV infection includes people requiring multiple blood/blood product transfusions, dialysis patients, injected drug users, household/sexual contacts of HBV infected, individuals with multiple sexual partners, individuals at occupational risk of exposure to HBV and inmates

of custodial institutions [3] and for HCV these include IV/intranasal drug users, recipients of multiple transfusions, sexual partners of HCV infected, people with HIV/other STIs, children born to HCV positive mothers, prisoners/previously incarcerated persons and people with tattoos/piercings [4]

Several of these high risk groups were present among HBsAg positives which included people requiring multiple transfusions, IV drug users, dialysis patients, house hold/close contacts of HBV infected and among HCV antibody positives those requiring multiple transfusions and people with HIV/any other STI were the most commonly identified. However, since only a small number of request forms had data on this regard it is difficult to get a comprehensive picture of the distribution of positives among risk groups.

Furthermore, analysis of the clinical presentations/indication for testing in the HBsAg and HCV antibodies revealed that majority were HBV chronic carriers and patients needing multiple transfusions respectively. However, some of the positives were form screening samples (screening before surgical procedure, antenatal screening, and medical screening) and HBV and HCV serology were positive in them as well, expressing the fact that the infection is not necessarily confined to high risk groups in the country and emphasizing the need for sero-prevalence studies based on the general population as well.

5. Conclusion

Sri-Lanka has a low prevalence for HBV infection and intermediate prevalence for HCV infection. The study confirmed the disease burden in the country and the need to strengthen and extend effective preventive measures to achieve the WHO declared elimination goals in Sri-Lanka by the year 2030.

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