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## **Original Research Article**

# Study of seroprevalence of hepatitis a and e viruses in patients of acute viral hepatitis in a tertiary care centre in sub himalayan region

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## ARTICLE INFO

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| Introduction: Acute viral hepatitis is a systemic infection affecting the liver predominantly. It is common |
|-------------------------------------------------------------------------------------------------------------|
| health problem in areas where safe drinking water is not universally available. In recent times we have     |
| witnessed frequent outbreaks as a result of that.                                                           |
| Aim: To estimate sero-positivity of Hepatitis A and E viruses in indoor patients of hepatitis, presenting   |
| with jaundice using ELISA.                                                                                  |
| <br>Settings and Design: Blood samples and clinical information were collected from cases of jaundice and   |
| referred to VRDL lab for analysis over a period of 1 year.                                                  |
| Materials and Methods: Samples were tested for anti -HAV immunoglobulin M (IgM) and anti -HEV               |
| immunoglobulin M (IgM).                                                                                     |
| Statistical Analysis: Data was entered into a pre-structured data entry form, on Microsoft Excel sheet.     |
| Results: The viral etiology was confirmed in 236 (41.1%) cases while in 337 (58.8%) cases no hepatitis      |
| virus could be detected, hepatitis E virus were found to be maximum in 45.76% cases, followed by HAV        |
| (39.83% cases) and while in 14.41% cases no hepatitis virus could be detected.                              |
| Conclusion: Routine basic microbiological analysis of drinking water should be carried out by assaying      |
| the presence of Escherichia coli by the multiple tube fermentation technique.                               |
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|                                                                                                             |

1. Introduction

Acute viral hepatitis is a systemic infection affecting the liver predominantly. ICMR-National Institute of Virology (NIV), Pune, received two batches of water samples from Shimla in January 2016 to test for the presence of enterically transmitted hepatitis viruses. Subsequently, 57 icterus patients were tested for various markers of hepatotropic viruses, i.e. anti-HEV IgM/IgG, anti-hepatitis A virus (anti-HAV) IgM/IgG antibodies and HEV RNA.<sup>1</sup> In view of the problem, the study was planned to highlight the viral agents responsible for causing hepatitis in patients presenting with jaundice in Indira Gandhi Medical College Shimla,India and to develop strategies for surveillance of preventable viral hepatitis at community level.

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## 2. Materials and Methods

## 2.1. Study area

The study was conducted in the Department of Microbiology in collaboration with the Department of Medicine and Pediatrics IGMC Shimla, Himachal Pradesh over a period of one year from 1st July 2016 to 30th June 2017.

#### 2.2. Study unit

Patients admitted to a tertiary care hospital with clinical suspicion of viral hepatitis presenting with clinical jaundice were included in the study.

## 2.3. Study design

It was a cross-sectional observational prospective hospital based study.

## 2.4. Inclusion criteria

Patients admitted to the Department of Medicine and Pediatrics at IGMC Shimla with viral hepatitis presenting with jaundice were included in the study.

## 2.5. Exclusion criteria

Known alcoholic patients on hepatotoxic drugs (e.g acetaminophen) and patient scrub typhus positive for IgM ELISA were excluded from the study.

## 2.6. Consent

Ethical clearance was obtained and informed consent taken.

## 2.7. Sample collection

Approximately 5 ml blood samples were collected aseptically from all cases. The venipuncture site was cleaned with soap and water, rinsed with sterile water and 1-2% tincture iodine or povidone iodine was applied and allowed to dry for 1-2 min (povidone-iodine) or 30 seconds (tincture of iodine). The tincture of iodine was removed with 70% alcohol. The blood was collected in sterile serum tubes. The tubes were then transported to the laboratory and kept on the rack and allowed to clot. The serum was separated.

## 2.8. Laboratory procedure

Serum was assessed for

- 1. anti HAV immunoglobulin M (IgM)
- 2. anti-HEV IgM antibodies

All analyses were performed using commercial kits based on the enzyme-linked immunosorbent assay (DSI,SRL Italy) as per the manufacturer's instructions.

## 3. Results and Discussion

A total of 573 serum samples from patients with clinical suspicion of jaundice were tested in the VRDL, IGMC Shimla out of which 236 patients serum were tested positive for viral hepatitis.

The viral etiology was confirmed in 236 (41.1%) cases while in 337 (58.8%) cases no hepatitis virus could be detected. Hepatitis E virus was found in the maximum number of cases (45.76% cases), followed by HAV (39.83% cases) and in 14.41% of the rest of the virus.

Similarly, a study conducted by Ganju et al<sup>2</sup> reported anti HAV antibodies in 37.86% cases, in contrast Jain et al<sup>3</sup> revealed that HAV accounts for 26.96% of viral hepatitis. In a study of bacteriological assessment of drinking water sources in and around Shimla city by Bhagra et al<sup>4</sup> revealed 11.74% of the water samples as unsatisfactory and not fit for human consumption

Out of 236 patients in the study 47(19.9%) were children in the age group of 4-18 yrs and 189(80.08%) were adults. Most common age group affected in Hepatitis A was 21-30 yrs followed by 31-40 yrs (17.02%) Whereas in Hepatitis E 41-50 yrs was the age group affected as shown in Table 1.

Studies conducted in the 2000s observed that nearly 90% of adolescents, adults, and most children acquired immunity to HAV infection in their preschool years. However, recent studies have indicated a shift in the epidemiology of HAV infection over the past decade.

With improvement in the socio-economic conditions of the communities, a shift in the age of acquiring HAV infection has been seen from childhood to older age groups in India and globally5. When compared with the seroprevalence in different cities among different age stratifications over a period of 10–15 years, a significant decline amongst all age groups of approximately 30% was noted. This difference in the number of HAV-seropositive cases over age clearly demonstrates a shift in epidemiology. This may be due to an increase in non-immune subjects over the years, resulting in changing epidemiology that might lead to a profound impact on the magnitude and the severity of the disease. This difference in anti-HAV prevalence could have been related to educational status and toilet facility.

It was observed that out of 45.76% cases HEV was most common in the age group 21-30 yrs (23.21%) as shown in Table 1. Co infection with HAV and HEV showed a similar trend. Co infection with HAV and HEV was seen in 25 patients. Incidence of HAV infection alone was higher in females(69.15%) compared to male(30.85%). In contrast Jain, et al<sup>3</sup> 2013 observed higher in Males (62.54%) as compared to females (37.45%). Incidence of HAV infection alone was higher in female (69.15%)compared to male(30.85%)

In this study group of clinically diagnosed patients, hepatitis A was more prevalent in the month of March (15.96%) followed by August, June and September(12.7%)

|                   | 0-10 | 11-20 | 21-30 | 31-40 | 41-50 | 51-60 | 61-70 |  |  |
|-------------------|------|-------|-------|-------|-------|-------|-------|--|--|
| Hepatitis A       | 7    | 8     | 13    | 10    | 8     | 4     | 6     |  |  |
| Hepatitis E       | 2    | 12    | 12    | 14    | 17    | 14    | 6     |  |  |
| Hepatitis A and E | 1    | 5     | 4     | 4     | 1     | 4     | 3     |  |  |

Table 1: Hepatitis A and E in different age groups

which may be attributed to flash floods during the rainy season and dry intervals in between. Jain et al<sup>3</sup> reported the maximum cases in the month from February through June (i.e autumn and summer seasons). Whereas Bhagra S et al<sup>4</sup> studied the seasonal trend more during the winter season (19 January to 31 March), summers from (1 April to 15 June) and monsoons (16 June to 31 July). In this study, 25(26.60%) were co-infection with hepatitis A virus and hepatitis E Chobe6 et al reported HAV outbreak in Shimla, the cases were reported from the areas getting water supply from Ashwani Khud water supply system. This water purification system received water from a natural stream in which treated sewage water was let into 4 km upstream the collection point for one year. Inadequate sewage management has played an important role in most waterborne outbreaks, which have generally involved contaminated groundwater obtained from wells or rivers and drinking water. The results of this present epidemiological study showed an overall high endemicity of HEV infection 47.58% and a significantly lower seroprevalence in the age groups <10 years and in 61-70 years. These differences might be due to either varying environmental conditions in different geographical areas and due to the prevailing outbreak in the last few months as reported by Bhagra et  $al.^4$ 



Fig. 1:

In this study group of clinically diagnosed patient, hepatitis E was more prevalent is month of August 27(25%) followed by September and October 16.67% and 13.8% as shown in Figure 2. Malathi et al.<sup>5</sup> (1998) reported increase in number of cases in month of February 1995 to January

1996. This may be due to inadequate sewage management which play an important role in most water-borne outbreaks generally contaminating the wells or rivers and drinking water.

In this study HAV 42.5% patients presented with yellowish discoloration of urine. The number of patients presenting with fever, loss of appetite, vomiting, pain in abdomen, diarrhoea were 61 (64.89%), 71 (75.53%), 52 (55.32%), 66 (70.21%), and 8(8.51%) respectively. Pallor was seen in 29(30.85%) and hepatomegaly was present in 13 (13.83%) patients. Encephalopathy was observed in 1(1.06%) patients as shown in Figure 2.



Fig. 2:

In this study HEV 71 (65.6%) patients presented with yellowish discoloration of urine. The number of patients presenting with fever, loss of appetite, vomiting, pain in abdomen, diarrhoea were, 71 (71.30%), 71 (75.53%), 55 (50.93%), and 57 (52.78%), 11(10.19%) respectively. Hepatomegaly was present in 11 (10.64%) patients. Encephalopathy was observed in 4(3.70%) patients as shown in Figure 2.

In this study, there was a history of travel to Shimla, 56.48% cases were resident of Shimla out of which 13.12% had a history of travel to endemic areas. In a study done by Bhagra et al<sup>4</sup> an outbreak of jaundice in Shimla which commenced in January 2016, confirmed to be due to Hep E virus by a team from NIV (National Institute of Virology), Pune (11.74%) were unsatisfactory i.e. unfit for drinking. This could be due to the unsatisfactory reports confirmed by this study, that maximum number of cases were of HEV.

In this study, the source of drinking water in 49(45.37%) people was tap water followed by boudi water in 39(36.11%) patients. In a study done in Shimla by Bhagra

et al<sup>4</sup> 26 (2.3%) public tap water was unsatisfactory which could be the reason for the spread of the virus as in Shimla water is supplied through seven surfaces according to Bhagra et al.<sup>4</sup> Ashwani WTP was contaminated with sewage effluent from Malyana and Dhalli STP (sewerage treatment plants). This lead to an outbreak of jaundice. In this study, the prevalence of HEV was more in Shimla 61 (56.48%) cases positive followed by Mandi 8(16.67%) cases.

In this study a question was asked to all the patients in the study group that, is the water you are drinking is safe for drinking, 42.3% peoples drinking either tap, water, water from step well or from hand pump were of the opinion that the water they are drinking is fit for drinking. This indicated that awareness and education has an impact on the water hygiene and sanitary habits of the population, irrespective of socioeconomic condition. Economic conditions and drinking water source did not show a significant effect on seroprevalence.

## 4. Conclusion

Safe drinking water for all is one of the major challenges of the 21st century. Routine basic microbiological analysis of drinking water should be carried out by assaying the presence of Escherichia coli by the multiple tube fermentation technique. Microbial contamination of drinking water has adverse health consequences so its control and prevention is of paramount importance and can never be compromised. Active intervention from public health officials and the health department along with raising people's awareness regarding water hygiene are required for improving the quality of drinking water. This study highlights that both HAV and HEV were prevalent in the area, which were transmitted by fecal-oral route, thus preventable. Provision of safe drinking water and safe sewage disposal systems are high priority besides and integration of hepatitis A vaccine into the existing national immunization schedule is recommended. Periodic surveillance of HAV/HEV is recommended to describe the epidemiological pattern in different geographic areas.

## 5. Source of Funding

None.

## 6. Conflict of Interest

The authors declare no conflict of interest.

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