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Title

Author

Abstract
Background: *In developing countries like India Hepatitis A (HAV) and hepatitis E (HEV) viruses are major concern for public health problems. These viruses are enterically transmitted through faeco-oral route and causes infection ranging from asymptomatic infection, usually in children, to acute viral hepatitis (AVH) of varying severity in adults.*
Aim & Objectives: *This study endeavors Seropositivity of Antibodies Against Hepatitis A and E*
- *This study is aimed at detection of IgM antibodies of Hepatitis A and E*
- *To study the age, gender and area prevalence of Hepatitis A and E*

Materials And Methods:
This is a observational study performed at Guntur Medical College, Guntur from June 2023 for 7 months.
- *The serum samples will be collected from patients suffering with fever and Jaundice in various departments of Government general hospital and tested at SL VRDL Department of Microbiology for Hepatitis A and E using enzyme linked immunosorbent assays (ELISA).*

Results: *A total of 548 patients' sample were tested for Hepatitis A and E. Out of which 247(45%) were Sero-positive for Hepatitis A and in Hepatitis E 35(6.8%) were positive. Among 247 hepatitis A positive cases,147(59.5%) were Males and 100(40.4%) were Females. Among 35 hepatitis E positive cases,18(51.4%) were Males and 17(40.4%) were Females and prevalence rate isseen more in urban areas.*
Conclusion: *For hepatitis A and hepatitis E better coordinated public health effort around this time can help contain the seasonal cases to some extent. an increasing trend of the number of cases reporting to the hospital, warrants the active community-based surveillance to assess the incidence of HAV and HEVin children and adults in this region. A long-term, continuous sero surveillance for presence of this viruses is important to ascertain the utility of the vaccine for its prevention.*
Keywords: *Elisa, Faeco Oral, Seropositivity,*

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I. Introduction

Hepatitis A and E are both viruses are predominantly enterically transmitted through faeco-oral route. Both HAV and HEV are non-enveloped icosahedral viruses. The lack of a lipid envelope offers both viruses a significant advantage in terms of their ability to spread in the environment, as demonstrated by the foodborne and waterborne outbreaks, which are synonymous with both hepatitis A and E

Hepatitis A is a vaccine-preventable disease but the vaccine has not been deployed in India as more than 80% of children by the age of 10 years develop antibodies as a result of natural infection and since the disease is often clinically insignificant in this age group. It is to note that more than half of the world's population practicing defecation in the open is residing in India.. To add to this, there has been an emphasis on the promotion of increased sanitary infrastructure by India under the Swachh Bharat (Clean India) mission since 2014.

HEV on the other hand is known to cause infection in adult population as compared to children, with a greater predilection to cause outbreaks in the community as compared to HAV. It is also documented to cause severe disease in pregnant females leading to increased mortality and pregnancy-related complications.There is evidence in the literature regarding this virus still being a public health menace in industrialized countries as well.

In India the mode of surveillance till date has been outbreak-oriented where the weekly numbers are analyzed by the Integrated Disease Surveillance Project, depending upon the geographical area. The National Viral Hepatitis Control Program (NVHCP), launched in July 2018, intends to address the public health problem caused by these viruses and it aims to substantially reduce the risk, morbidity, and mortality associated with HAV and HEV by 2030.

There are limited long-term studies from India regarding the extent of the disease burden of these two viruses. Considering the diverse socio-economic and demographic factors in a vast country like India, coupled with recent improvements in the sanitation infrastructure under clean India Mission 2014, it is important to study the long-term trends of HAV and HEV infections.

II. Materials And Methods

Sample collection

Whole blood (4-5 ml) was collected from patients with acute fever and jaundice by venipuncture into red capped plain vacutainer under strict aseptic condition.

To separate the serum, the sample were kept at room temperature till clot formation and centrifuged at 3000 rpm for 10 min.

Sample processing

Blood collected in the plain vial was allowed to clot and after centrifugation serum was separated for detection of for igm antibody to hepatitis A and E virus.

Elisa Kit For IGM Antibody To Hepatitis A Virus.

HAV IgM – ELISA

Test procedure

- The procedure was done as per the instruction given in the kit as follows
- Select the sample to be tested. write down the protocol on ELISA sheet provided with each kit.
- Bring ELISA kit for antibody IgM to hepatitis A virus (all reagents), and samples to room temperature before use (approximately 30 minutes).
- Dilute concentrated wash buffer 1:19 with ddH2O.
- Dilute the sample (1:1000) with physiological saline.
- For each test, set one blank,two positive and 3 negative controls. Add 100 microlitres positive and negative control serum wells respectively.
- Add 100 micro litres diluted samples into other test wells.
- Cover wells with seal paper, then incubate 30 minutes at 37 degree Celsius.
- Discard the liquid in all wells and fill the wells with wash solution.lay aside for 15 seconds,discard the liquid in all wells and fill the wells with wash solution.Repeat 5 times and dry wells after lst wash.
- Add 50 microlitres conjugate HAV-Ag in each well except the blank.
- Add 50 microlitres enzyme conjugate in each well except the blank.
- Cover wells with seal paper,then incubate 30 minutes at 37 degree Celsius.
- Repeat step 7
- Add 50 microlitres substrate Aand B respectively to each well,mix gently protected from light and incubate 15 minutes at 37 degree Celsius.
- Add 50 microlitres of stop solution into each well to stop the reaction ,including blank well.
- Measure the absorbance at450nm against the blank,or measure the absorbance at 450nm/630-690nm.

HEV IgM – ELISA

1.Elisa Kit For Igm Antibody To Hepatitis E Virus.

Test procedure

The procedure was done as per the instruction given in the kit as follows

- Select the sample to be tested. write down the protocol on ELISA sheet provided with each kit.
- Bring ELISA kit for antibody IgM to hepatitis E virus (all reagents), and samples to room temperature before use (approximately 30 minutes).
- Dilute concentrated wash buffer 1:19 with ddH2O.
- Dilute the sample (1:1000) with physiological saline.
- For each test ,set one blank,two positive and 3 negative controls.Add 100 microlitres positive and negative control serum wells respectively.
- Add 100 micro litres diluted samples into other test wells.
- Cover wells with seal paper ,then incubate 30 minutes at 37 degree Celsius.
- Discard the liquid in all wells and fill the wells with wash solution.lay aside for 15 seconds,discard the liquid in all wells and fill the wells with wash solution.Repeat 5 times and dry wells after lst wash.
- Add 50 microlitres conjugate HEV-Ag in each well except the blank.
- Add 50 microlitres enzyme conjugate in each well except the blank.
- Cover wells with seal paper,then incubate 30 minutes at 37 degree Celsius.
- Repeat step 7
- Add 50 microlitres substrate Aand B respectively to each well,mix gently protected from light and incubate 15 minutes at 37 degree Celsius.
- Add 50 microlitres of stop solution into each well to stop the reaction ,including blank well.
- Measure the absorbance at450nm against the blank,or measure the absorbance at 450nm/630-690nm.

III. Results:

A total of 548 patients sample were tested for Hepatitis A and E. Out of which 247(45%) were Sero-positive for Hepatitis A and in Hepatitis E 35(6.8%) were positive.

Prevalence Of Hepatitis A And E

S NO	NAME OF VIRUS	TOTAL SAMPLES	TOTAL POSITIVES
1	HEPATITIS A	548	247 (45.07%)
2	HEPATITIS E	548	35 (6.4%)

Sexwise Distribution

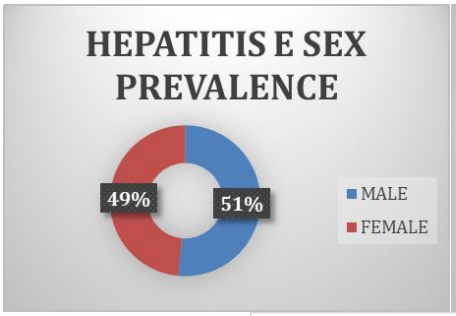
S NO	NAME OF VIRUS	MALES	FEMALES
1	HEPATITIS A	147 (59.5%)	100(40.4%)
2	HEPATITIS E	18 (51.4%)	17 (48.5%)

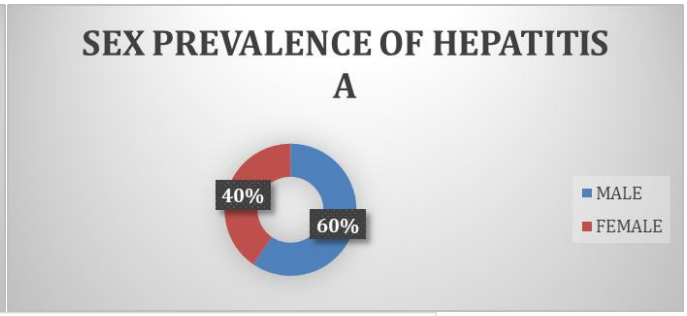
Agewise Distribution:

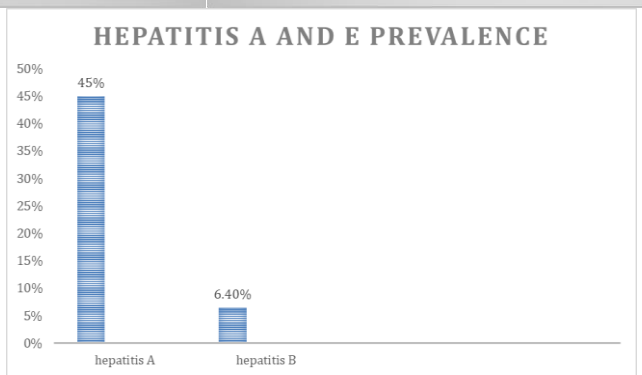
AGE	HEPATITIS A	HEPATITIS E
0 TO 18YRS	184 (74.4%)	30 (85.7%)
19 YRSTO 45YRS	44 (17.8%)	4 (11.4%)
46YRS TO 60YRS	15 (6.07%)	1 (2.8%)
>60YRS	4 (1.6%)	0

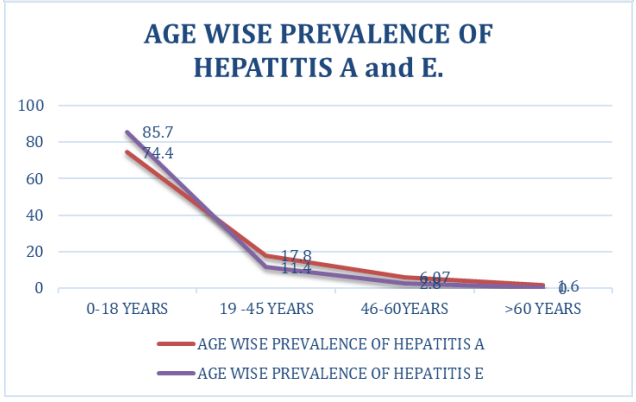
Area Wise Distribution:

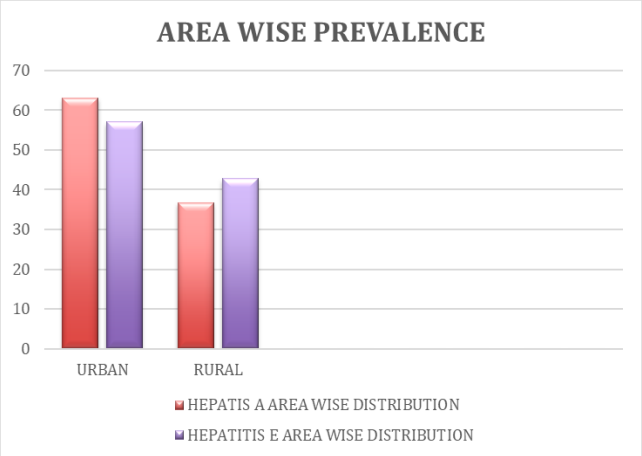
AREA	HEPATITIS A	HEPATITIS E
URBAN	156 (63.1%)	20 (57.1%)
RURAL	91 (36.8%)	15 (42.8%)











IV. Discussion:

In our present study the prevalence of hepatitis A is 45.07%, where highest is seen in Bansal etal with 16.9% and lowest prevalence is seen in palewar etal with 6.7%

The prevalence of hepatitis E is 6.4% and it is corelated with the palewar etal whose prevalence is 8.5% and highest is seen in Kalita etal with 28% and least seen in palewar etal.

The sex prevalence of hepatitis A in our study (male 59.5&female 40.4%) corelates with A Joon (male 68 &female 31%). The highest male sex prevalence seen Iin A Joon etal and lowest male prevalence is seen in MurhekarMV etal , the highest female sex prevalence seen in MurhekarMV etal.

The sex prevalence of hepatitis E in our study is high in male (male 51.4&female 48.5%) corelates with A Joon (male 68 &female 31%) and least is seen with murhekarMV etal age wise prevalence of hepatitis A in our study corelates with palewar etal (less than 18 years),in study of murhekarMV etal age wise prevalence of hepatitis A is more in less than 9 years and in AJoon etal the age wise prevalence is more in between 21-25 years. age wise prevalence of hepatitis E in our study corelates with palewar etal with 20 years and murhekar MV etal shows age prevalence between 20-29 years A Joon etal shows between 21-25 years .

V. Conclusion

The HAV seroprevalence (45.07%)is higher than HEV(6.4%) . The HAV predominating in relatively Young patients (age group 0-18 years) compared to Older subjects in HAV infections with male prepondarence (59.5%), which is seen more in Urban than rural . Therefore the present study emphasizes the importance of screening hepatitis viral markers (A, E) for early diagnosis and curtailment of outbreaks and epidemics by the public health sector reducing morbidity and mortality. With a faeco-oral route of transmission, periodic surveillance, especially in monsoon and post-monsoon, is of utmost importance for early diagnosis and curtailment of outbreaks and epidemics by the public health sectors through proper sanitization, hygiene, and public awareness. Considering the high seroprevalence of anti-HAV antibodies in the general population, mass immunization with the hepatitis A vaccine may not be cost-effective in a country like India, however, its use in risk populations, like chronic liver disease patients, during the onset of outbreaks and epidemics, travelers to endemic areas, and in younger children, should be considered

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