

Assessment of the Prevalence of hepatitis C among pregnant females

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Abstract

Background: Hepatitis C virus (HCV) is a major causative agent of parenterally acquired hepatitis. Vertical transmission of HCV occurs from mother to infant during pregnancy, delivery, or neonatal period. Hepatitis C infection among pregnant females can adversely affect both mother and fetus, leading to chronic infection in infants who later develop liver cirrhosis and hepatocellular carcinoma. **Aim:** The aim of the study was to evaluate the prevalence of hepatitis C among pregnant females attending antenatal clinic. **Methods:** A hospital based cross-sectional study was done over a period of seven months from June to December 2020. A total of 550 pregnant females were included in the study who attended Antenatal Clinic for routine check-up and whose blood samples were sent to Microbiology Laboratory for screening of anti-HCV antibodies by Enzyme linked Immunosorbent Assay (ELISA) method. **Results:** Out of 550 pregnant females tested, 3 were found to be reactive to anti-HCV antibodies by ELISA; hence the prevalence was found to be 0.5%. All 3 seropositive females belonged to age group 26-35 years, were indoor patients (IPD) and belonged to lower socio-economic class. Majority of seropositive females belonged to urban areas (66.7%, 2/3) as compared to those from rural areas (33.3%, 1/3). **Conclusions:** Although, prevalence of hepatitis C was found to be less among pregnant females attending ANC of this institute, still routine screening of all pregnant females for anti-HCV antibodies is highly recommended, for timely detection of disease in mothers and prevention of its transmission to infants.

Keywords: Enzyme-linked immunosorbent assay, Hepatitis C, Pregnant females, Prevalence

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Introduction

Hepatitis C virus (HCV) is the major etiological agents of parenteral acquired hepatitis[1]. Hepatitis C is found worldwide. The most affected regions are the World Health Organization (WHO) Eastern Mediterranean Region and the WHO European region, with an estimated prevalence in 2015 of 2.3% and 1.5% respectively.

Prevalence of HCV infection in other WHO regions varies from 0.5% to 1.0%.² Increasing incidence of hepatitis C infection among pregnant women affecting both mother and fetus during pregnancy have been reported and this can lead to chronic infection in the infant who later develop liver cirrhosis and hepatocellular carcinoma[3] More than 8% of pregnant women are infected with HCV worldwide.⁴ Studies suggest prevalence of HCV among pregnant females to be 0.6% - 2.4%, with overall mother to infant transmission varying from 8% to 15%[5,6]

HCV is a single stranded, enveloped RNA virus with a genome organized as 1 nm long open reading frame. Hepatitis C virus has some structural proteins (Core, E1 and E2) which are at the end of the genome and are followed by the nonstructural proteins[7]. Genetic diversity of HCV exhibits by 6 main genotypes and also hypervariable regions within the E2 protein[8].

Vertical transmission of HCV occurs from mother to infant during pregnancy, delivery, or neonatal period, resulting in hepatitis C infection in children[5,9] Not every transmission event result in

chronic infection in infants[10- 12]. Hepatitis C can cause acute or chronic hepatitis. Mostly acute hepatitis C is asymptomatic and hence individual is unaware of the underlying infection[13]. Almost 20%-30% of infected cases automatically resolve the virus and remaining 75%-80% of cases progress to chronic hepatitis. Approximately 10%-25% of people with chronic hepatitis C develop cirrhosis and hepatocellular carcinoma, a process that usually takes 20-30 years[14,15]. In Central Europe the seroprevalence of anti-HCV antibodies was estimated to be 2.4% and in Eastern Europe 2.9% and in Western Europe 2.4%. [16]

Countries with high prevalence of HCV infection include Egypt >10% or African countries > 3%.¹⁷ Countries with lowest seroprevalence of HCV infection include Netherland, Scandinavia ≤0.5%.¹⁸ In pregnant women seroprevalence of anti-HCV antibody is around 1.03% in India[19] With rising incidence of HCV a significant number of pregnant women are affected with HCV, and there is little research regarding the impact of HCV on pregnancy out-come in India[20]

Hence, the present study was done to evaluate the prevalence of Hepatitis C among pregnant females attending for Antenatal check up at our Institute.

Methods

This hospital based Prospective cross-sectional study was conducted at Department Of Gynaecology And Department Of Microbiology, at Vardhman Institute of Medical Sciences, Pawapuri. The study was conducted over a period of 07 months time from duration June 2020 – December 2020. The study was approved by institutional research and ethical committee. An informed and written consent was taken from all the participants prior to the commencement of the study.

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This study determined the prevalence of Hepatitis C among pregnant females attending our hospital for Antenatal check up. A pre-designed questionnaire was used to determine the socio-demographic profile of the enrolled patients.

Inclusion criteria: Apparently healthy pregnant females (Outpatient, OP and indoor patients, IP) attending Antenatal clinic who were screened for hepatitis C and who gave their consent were included in this study.

Exclusion criteria: Patients who did not give their consent were excluded.

A total of 550 pregnant females whose blood samples were sent to Microbiology laboratory for anti-HCV antibodies testing were included in the present study.

3ml of venous blood withdrawn aseptically from each pregnant female in a well labeled plain vacutainer tube was received in Microbiology Laboratory. The blood was allowed to clot followed by centrifugation of the tube at 3000 rpm for 15 min to separate serum. The sera was then tested for anti-HCV antibodies by using 3rd generation HCV Microlisa an enzyme linked immunosorbent assay (ELISA) method (J. Mitra and Company Private Limited, India). The HCV Microlisa is based on detection of host generated anti-HCV antibodies to viral proteins (Core E1, E2 and Non-Structural NS3, NS4 and NS5 antigens) coated into the microwells. The test was performed according to manufacturer’s instruction. 100 µl negative control, 100 µl positive control, 100 µl sample diluent and 10 µl samples were added in the respective wells. The plate was then

covered and incubated in an incubator at 37°C±2°C for 30 minutes followed by washing with working wash solution. Then 100 µl working conjugate was added in each well. The plate was again covered and incubated in an incubator at 37°C±2°C for 30 minutes. The plate was then washed with working wash solution followed by the addition of 100 µl working substrate solution in all the wells. The plate was then covered and incubated at room temperature (20-30°C) for 30 minutes in dark. Finally, 100 µl stop solution was added to each well and the absorbance was read at 450 nm in an ELISA reader. The Cut-off value was calculated by multiplying 0.23 with the mean absorbance of Positive control. Samples with absorbance value less than the cut-off value were considered as non-reactive, whereas, those with absorbance value equal to or greater than the cut-off value were considered as reactive by the criteria of HCV Microlisa.

Statistical analysis

The collected data was analyzed using SPSS Data Editor Software version 11.0. Chi-square test and Fisher’s exact test were performed for categorical variables and p value≤0.05 were considered statistically significant.

Results

A total of 550 pregnant females included in the study were screened for presence of anti-HCV antibodies. Of these 3 pregnant females were found to be reactive by HCV Microlisa (ELISA), giving the seroprevalence of Hepatitis C to be 0.5% as shown in (Table1).

Table 1: Distribution of pregnant females according to the results of HCV Microlisa (N = 550).

Result of HCV Microlisa	Number of pregnant females (N)	Percentage (%)
Reactive	3	0.5%
Non-reactive	547	99.5%
Total	550	100%

It was found that majority of pregnant females included in this study belonged to age group 15-25 years (45.3%, 249/550), followed by age group 26-35 years (43.3%, 238/550) and least number belonged

to age group 36-45 years (11.4%, 63/550). However, all the 3(100%) reactive female patients belonged to age group 26-35 years, as depicted in (Table 2).

Table 2: Distribution of pregnant females according to their age group and reactivity to HCV Microlisa (N = 550).

Age group (in yrs)	Result of HCV Microlisa		Total
	Reactive N (%)	Non-reactive N (%)	
15-25	0 (0%)	249(100%)	249(100%)
26-35	3 (1.3%)	235 (98.7%)	238(100%)
36-45	0 (0%)	63 (100%)	63 (100%)
Total	3 (0.5%)	547 (95.5%)	550(100%)

N = Number of pregnant females.

In this study, although, majority of pregnant females belonged to rural areas (74%, 407/550) as compared to those from urban areas (26%, 143/550), higher number of reactive patients belonged to

urban areas (66.7%, 2/3) as compared to those from rural areas (33.3%, 1/3). However, this difference was not found to be statistically significant (p = 0.11) (Table 3).

Table 3: Distribution of pregnant females according to their residence and reactivity to HCV Microlisa (N = 550)

Residence	Result of HCV Microlisa		Total	Chi-Square(χ ²) and *p value
	Reactive N (%)	Non-reactive N (%)		
Urban	2(1.4%)	141(98.6%)	143(100%)	χ ² = 2.52 df = 1 p = 0.11
Rural	1(0.2%)	406(99.8%)	407(100%)	
Total	3(0.5%)	547(99.5%)	550(100%)	

* p<0.05 was considered as statistically significant. df = degree of freedom, N = Number of pregnant females.

Table 4 shows, majority of pregnant females belonged to indoor patient department or IPD (66.7%, 367/550) as compared to those who belonged to outpatient department or OPD (33.3%, 183/550).

Also, it was found that all 3 reactive patients belonged to IPD. However, this finding was not found to be statistically significant (p = 0.554).

Table 4: Distribution of pregnant females according to their registration status and reactivity to HCV Microlisa (N = 550)

Registration status	Result of HCV Microlisa		Total	Fisher's Exact test
	Reactive N (%)	Non-reactive N (%)		
OPD	0 (0%)	183 (100%)	183 (100%)	p = 0.554
IPD	3 (0.8%)	364 (99.2%)	367 (100%)	
Total	3 (0.5%)	547 (99.5%)	550 (100%)	

* p<0.05 was considered as statistically significant. N = Number of pregnant females. OPD = Outpatient department. IPD = Indoor patient department.

Author found that majority of pregnant females enrolled in this study belonged to lower socio-economic class (68.4%, 376/550), followed by those belonging to middle class (30.0%, 165/550) and least from

upper class (1.6%, 9/550). All the 3 reactive females belonged to lower class, as shown in (Table 5).

Table 5: Distribution of pregnant females according to their socio-economic status and reactivity to HCV Microlisa (N = 550).

Socio economic status	Result of HCV Microlisa		Total
	Reactive N (%)	Non-reactive N (%)	
Upper class	0(0%)	9(100%)	9(100%)
Middle class	0(0%)	165(100%)	165(100%)
Lower class	3(0.8%)	373(99.2%)	376(100%)
Total	3(0.5%)	547(99.5%)	550(100%)

N = Number of pregnant females.

Discussion

HCV infection among pregnant women vary worldwide, from 1% to 2.5% in the United States and Europe to more than 10% in some sub-Saharan countries.21-23 In India the overall anti-HCV positivity varies from 0.6% to 1.4%.24 The epidemiology of HCV varies among countries and the reported prevalence of HCV in pregnant women has not been extensively studied, due to lack of screening of infection and the lack of preventive measures for mother- to-child transmission[25].In this study, 550 pregnant females were screened for anti-HCV antibodies, and author found that 3 pregnant females were reactive to HCV by ELISA test, hence the prevalence of HCV infection in pregnant females was 0.5%. A study done from Chennai reported prevalence of HCV among pregnant women to be 0.6%[26] Another study from Uttarakhand, also reported seroprevalence of hepatitis C to be 0.89% among pregnant women[27] Both these studies corroborate this finding. However, in contrast to this finding, a study from Sangareddy, Telangana state reported very low prevalence (0.21%) of HCV infection among pregnant women[28] Similarly, studies from New Delhi and Punjab reported high prevalence of hepatitis C among pregnant women to be 1.03% and 2.8% respectively[20,29].This study found that majority of pregnant females belonged to age group 15-25 years (45.3%,249/550), followed by age group 26-35 years (43.3%,238/550), however, all the 3(100%) reactive female patients belonged to age group 26-35 years. A study from Sangareddy, Telangana state, reported that majority of seropositivity was seen among pregnant women belonging to age group 21-30 years (66.7%,2/3)[28] Another study from Faridkot, Punjab also reported higher seropositivity among pregnant women belonging to age group 21-25 years (45%, 18/40) and 26-30 years (30%, 12/40) [29] These findings were comparable to the present study.

In this study although, majority of pregnant females belonged to rural areas (74%, 407/550) as compared to those from urban areas (26%, 143/550), higher number of reactive patients belonged to urban areas (66.7%, 2/3) as compared to those from rural areas (33.3%, 1/3). In contrast to this finding, a study from Faridkot, Punjab reported higher seropositivity among pregnant women belonging to rural areas (72.5%, 29/40) as compared to those from urban areas (27.5%, 11/40)[29]

In the present study it was found that majority of pregnant females belonged to lower socio-economic class (68.4%, 376/550), followed by those belonging to middle class (30.0%, 165/550) and least from upper class (1.6%, 9/550). All the 3 reactive females belonged to

lower class. A study from Chennai also reported that all the 18 reactive pregnant women belonged to lower class and none belonged to upper middle class[26].

Conclusion

To conclude, although prevalence of Hepatitis C among pregnant females attending Antenatal clinic of our hospital was less, still routine screening of all pregnant females for anti-HCV antibodies is highly recommended, as early diagnosis of disease in antenatal period is helpful for proper patient management and initiation of treatment to prevent transmission of HCV to newborns. By screening of only at risk females one may miss significant proportion of pregnant women with HCV infection, as majority of pregnant women are asymptomatic, they might not be aware of their infection status and thus transmit their infection to newborns.

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