Original Article

Seropositivity of hepatitis B, hepatitis C, syphilis, and HIV in antenatal women in India

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Abstract

Introduction: The epidemiology of viral hepatitis during pregnancy is of paramount importance for health planners and program managers. Data on viral hepatitis during pregnancy are not readily available. This study was conducted to assess the extent of seropositivity of hepatitis B, hepatitis C, HIV, and syphilis in pregnant women and to re-evaluate the need for routine antenatal care screening.

Methodology: All samples were tested to detect HBsAg by enzyme linked immunosorbent assay (ELISA). Samples were tested to detect anti-HCV by ELISA. Samples were also tested for antibodies to Treponema Pallidum by qualitative rapid plasma reagine (RPR); finally, samples were tested for antibodies to HIV by three different methods as per Strategy III of the National AIDS Control Organization by using different systems of testing to establish a diagnosis of HIV.

Results: Seropositivity of hepatitis B was 2.9%, hepatitis C was 0.19%, syphilis was 0.48%, and HIV was 0.38%. Out of the 1038 samples, no co-infection was found between hepatitis B, hepatitis C, syphilis, or HIV.

Conclusion: The data from this study can help health professionals to treat antenatal patients more effectively. The data also reinforces the need for establishing effective prevention programs, which could lead to a reduction in the prevalence of HBV, HCV, syphilis, and HIV

Key words: antenatal women; hepatitis B; hepatitis C; HIV; seropositivity; syphilis

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Introduction

Hepatitis B virus is the most important causative agent of transfusion-associated hepatitis. Humans are the only reservoir of Hepatitis B virus (HBV). The significance of HBV infection during pregnancy derives through its potential to be transmitted vertically. Ten percent of infants born to women with acute HBV infection during the first trimester of pregnancy are HBsAg-positive at birth and80 to 90% of neonates become HBsAg-positive without prophylactic therapy if acute maternal infection develops during the third trimester of pregnancy [1].

According to Okada *et al*., 85% of neonatal HBV infections are caused due to intrapartum exposure to infectious blood and vaginal secretion, and the remaining 15% are caused by haematogenous transplacental viral spread. [2]

Little is known about hepatitis C virus (HCV) infection in pregnant women in India. The seroprevalence of anti-HCV antibody in the healthy general population of India was found to be 1.5 per cent each in 234 voluntary blood donors and 65 pregnant women [3]. Large scale studies on the estimates of the prevalence of HCV infection or the risk behavior of HCV infection in the low-risk Indian population are yet to be done. The current study is to assess the prevalence of HCV infection within an obstetric population attending our hospital.

Transmission of *T. pallidum* from a syphilitic woman to her fetus through the placenta may occur at any stage of pregnancy, but the lesions of congenital syphilis generally have their onset after the fourth month of gestation, when fetal immunologic competence begins to develop. This timing suggests that the pathogenesis of congenital syphilis depends on the immune response of the host rather than on a direct toxic effect of *T. pallidum*. The risk of infection of the fetus during untreated early maternal syphilis is estimated to be 75 to 95%, decreasing to approximately 35% for maternal syphilis of two years' duration [4].

HIV infection can be transmitted from an infected mother to her fetus during pregnancy, during delivery, or by breastfeeding. This is a highly potent form of HIV transmission in developing countries, where the proportion of infected women to infected men is 1:1.

Virology analysis of aborted fetuses indicates that HIV can be transmitted to the fetus as early as the first and second trimesters of pregnancy [4].

Methodology

This study was conducted to determine the prevalence of hepatitis B virus surface antigen (HBsAg), antibodies to hepatitis C virus, antibodies to *Treponema pallidum*, and antibodies against HIV virus among patients attending the antenatal clinic of P. D. U. Medical College and Hospital in Rajkot. Serum samples from 1038 cases were collected between February 2010 and May 2010. These samples were tested for hepatitis B (HbsAg), hepatitis C, syphilis, and HIV per Strategy III of the National AIDS Control Organization by using different test systems to establish diagnosis of HIV.

Five-mL blood samples were collected using a sterile plain vacutainer, and the serum was separated by centrifugation into sterile serum storage vials. Needles were destroyed using a needle destroyer and then discarded in a sharps box. Other contaminated materials were discarded in 1% hypochlorite solution.

Laboratory tests for HBsAg

The serum samples were checked for the presence of hepatitis B surface antigen (HBsAg) using Microscreen HBsAg ELISA (M/S Span Diagnostics Ltd, Surat, India), which is a direct noncompetitive solid phase enzyme immunoassay for the detection of HBsAg in serum or plasma.

Laboratory tests for HCV antibodies

The serum samples were checked for the presence of IgG antibodies to HCV using Innova HCV (M/S Span Diagnostics Ltd, Surat, India) test kit, a third-generation ELISA for the detection of antibodies against HCV in human serum or plasma. Micro wells were coated with HCV-specific recombinant antigens from the putative C-core (structural), E1 and E2 (envelop proteins), NS3, NS4, and NS5 (non-structural) regions of the HCV genome. The test was performed according to the manufacturer's instructions.

Laboratory diagnosis for syphilis

Serum samples from all donors were tested for the presence of treponemal antibodies using carbogen. The RPR syphilis screening test (Tulip Diagnostics Pvt Ltd, Goa, India) which is a macroscopic non-treponemal flocculation card test for the detection and quantitation of antilipoidal antibodies present in serum or plasma of syphilitic persons was used. Briefly, serum or plasma was mixed with the reagent and allowed to act for eight minutes. When antilipoidal antibodies were present in the specimen, they reacted with the reagent, forming visible black floccules. When antilipoidal antibodies were absent in the specimen, flocculation was not observed. The test was performed using the manufacturer's instructions.

Laboratory diagnosis for HIV

Antibodies to HIV (anti-HIV) were determined by dot immunoassay (CombAids HIV 1 + 2 Immunodot Test Kit, M/S Span Diagnostics Ltd, Surat, India), and positive results were confirmed by the test which employs lateral flow-immunochromatographic type assay line immunoassay (Pareekshak HIV 1/2 Triline card test, Bhat Biotech Pvt Ltd, Bangalore, India) and the HIV TRI-DOT test (J. Mitra & Co Pvt Ltd, New Delhi, India), which is a visual, rapid, sensitive and accurate immunoassay for the detection of HIV-1 and HIV-2 antibodies (IgG) in human serum or plasma using HIV-1 and HIV-2 antigens immobilized on a porous immunofiltration membrane.

Results

A total of 1038 samples from antenatal patients were screened for hepatitis B virus infection, hepatitis C virus infection, syphilis infection, and HIV infection as under:

1. Detection of HBsAg by ELISA test.
2. Detection of anti-HBC antibody by ELISA test.
3. Detection of antibody to T. pallidum by RPR test.
4. Detection of antibodies to HIV by three different methods, according to Strategy III of the National AIDS Control Organization, India).

The seroprevalence of HBsAg among antenatal cases was maximum in the 21-25 year age group (58.06%). There were 31 samples positive for HBsAg out of 1038 samples; hence the overall prevalence for HBsAg was 2.9%. (Table 1). The prevalence of HBsAg in the second trimester was the highest (45.16%), followed by the first (32.26%) and third trimester (22.58%) (Table 2).

There were two samples positive for anti-HCV out of 1038 samples, thus the overall prevalence for anti-HCV was 0.19% (Table 1). There were two samples seropositive for anti-HCV in the first and second trimesters (Table 2).

Prevalence of the T. pallidum antibody among antenatal cases was maximum in the 21-25 year age group (100%); all the positive cases were from this

Table-1: Hepatitis-B, Hepatitis-C, Syphilis and HIV among antenatal cases in various age groups.

		Age-groups(in years)					Total	Chi-square	P value
		17-20	21-25	26-30	31-35	>35			
Hep-B	Negative	230	554	189	30	4	1007	1.33	0.86
	Positive	8	18	5	0	0	31		
	Total	238	572	194	30	4	1038		
Hep-C	Negative	238	571	194	29	4	1036	16.24	0.003
	Positive	0	1	0	1	0	2		
	Total	238	572	194	30	4	1038		
Syphilis	Non-Reactive	238	567	194	30	4	1033	4.09	0.39
	Reactive	0	5	0	0	0	5		
	Total	238	572	194	30	4	1038		
HIV	Non-Reactive	238	571	191	30	4	1034	8.52	0.07
	Reactive	0	1	3	0	0	4		
	Total	238	572	194	30	4	1038		

Table-2: Hepatitis-B, Hepatitis-C, Syphilis and HIV among antenatal cases in various Trimesters.

		Trimester			Total	Chi-square	P value
		1st	2nd	3rd			
Hep-B	Negative	330	390	287	1007	1.43	0.49
	Positive	7	14	10	31		
	Total	337	404	297	1038		
Hep-C	Negative	336	403	297	1036	0.83	0.66
	Positive	1	1	0	2		
	Total	337	404	297	1038		
Syphilis	Non-Reactive	336	403	294	1033	2.43	0.29
	Reactive	1	1	3	5		
	Total	337	404	297	1038		
HIV	Non-Reactive	337	400	297	1034	6.30	0.04
	Reactive	0	4	0	4		
	Total	337	404	297	1038		

Table-3: Comparison of percentage of positive subjects of HBsAg among Antenatal cases

Sr. No.	Study	Total Sample	HBsAg +	percentage of positive subjects
1	Mittal et al[5]	850	54	6.3
2	Gill et al[6]	2000	100	5
3	Nayak et al[7]	8575	322	3.7
4	**Present Study**	1038	31	2.9
5	Panda et al[8]	8431	191	2.6
6	Sehgal et al[9]	4137	109	2.6
7	Gupta et al[10]	2337	58	2.5
8	Biswas et al[11]	1000	23	2.3

Table-4: Comparison of percentage of positive subjects of anti-HBC antibody among Antenatal cases

Sr. No	Study	Total Sample	percentage of positive subjects
1	Gangu et al[13]	250	0
2	**Present study**	1038	0.19
3	Rudrapathy et al[14]	3115	0.6
4	Ashok Kumar et al[15]	8130	1.03
5	Farhana Shaikh et al[16]	3020	3.44

Table-5:-Comparison of percentage of positive subjects of HIV antibody among antenatal cases

Sr. No.	Study	Total Sample	percentage of positive subjects
1	Mathur et al[20]	2550	1.86
2	M.Mustafa et al[21]	3602	1.1
3	Gupta et al[22]	3529	0.88
4	**Present study**	**1038**	**0.38**

age group. There were five samples positive for T. pallidum antibody out of 1038 samples;thusthe overall prevalence of the T. pallidum antibody was 0.48% (Table 1). Three samples from the third trimester were seropositive, while only one sample from the first and second trimesters each were seropositive (Table 2).

All samples were tested using qualitative rapid plasma reagin (RPR); reactive samples were retested with quantitative RPR to rule out biological false positive samples. All reactive qualitative RPR had a titer ≥1:8.

The prevalence of HIV was highest in the 26-30 year age group (75%), followed by the 21-25 year age group (25%). A total of four samples out of 1038 were positive for HIV; the overall prevalence for HIV was 0.38% (Table 1). HIV seroprevalence was highest in the second trimester (100%) (Table 2).

Out of the 1038 samples, no co-infection was found between hepatitis B, hepatitis C, syphilis, or HIV.

Discussion

This study showed an HBsAg prevalence rate of 2.9% among antenatal women, which is lower than the rates reported by Mittal et al. (6.3%) [5] and Gill et al. (5%) [6]. Our results are comparable to those of Nayak et al. (3.7%) [7], Panda et al. (2.6%) [8], Sehgal et al. (2.6%) [9], and Gupta et al. (2.5%) [10]. The results from our study is slightly higher than those reported by Biswas et al. (2.3%) (Table 3).

The strong possibility of vertical transmission lends importance to diagnosing acute or chronic HBV infection in pregnant women and justifies mandatory antepartum serum HBsAg screening. [12] Screening of HBsAg will reveal previously unsuspected chronic HBV infection in young, otherwise healthy, individuals. This screening has the added benefit of making it possible to refer such patients for appropriate antiviral therapy before significant liver damage and associated functional insufficiency are developed.

Large scale studies on the estimates of the prevalence of HCV infection and risk behavior of HCV infection in the Indian population are yet to undertaken. This study assessed the prevalence of HCV infection within antenatal women in PDU Hospital in Rajkot.

Of the 1038 samples included in this study, only two samples were positive for anti-HCV antibodies, with a prevalence of 0.19%, which is very low compared to the rates reported by Farhana et al. (3.44%) [16] and Ashok et al. (1.03%) [15], but similar to the rates reported by Rudrapathy et al. (0.6%) [14] and Gangu et al. (0%) [13] (Table 4). In our study, 50% of HCV-positive patients were in the 21-25 year age group, which is comparable to the findings of Rudrapathy et al.](52%) [14].

With a prevalence of HCV infection lower than the rest of the world, identification of HCV infection poses a greater public health problem. The modules

based on selective screening for high risk factor analysis will fail to identify over half of infected patients. Therefore, targeted screening is not sufficient, and universal screening would present cost constraints, especially in resource-poor countries.

The prevalence rate of syphilis in the present study (0.48%) was very low compared to the rate reported by Kebede et al. [18] (2.9%), and the rate reported by Gupta et al., [19] (1.47%). In India, available information indicates that the prevalence of maternal syphilis has remained at around 1.5% between 2003 and 2007 [17].

During the study period, 1038 pregnant women were screened for HIV, and 0.38% (n = 4) tested positive for HIV, which is slightly lower than the prevalence in antenatal women in India between 2009 and 2010 (0.49%) [23].

The findings in our study were lower compared to the findings of Mathur et al. (1.86%) [20], Mustafa et al. (1.1%) [21], and Gupta et al. (0.88%) [22] (Table 5).

Our study indicates a lower trend of HIV prevalence. Although our study population is not representative of the whole of India due to limitations to sample size in a single hospital study, the data illustrates the spread of HIV in pregnant mothers. This will directly lead to high perinatal transmission and a reciprocal increase in pediatric AIDS cases. Therefore, it may be recommended that even though the curative treatment for HIV is not available at present, we can minimize, if not prevent, pediatric HIV infection by early screening of pregnant mothers for HIV followed by perinatal short-term anti-retroviral therapy, safe delivery practices, and modified infant feeding.

Conclusion

This data can help health professionals to efficiently treat antenatal patients. The data also reinforces the need for effective prevention programs, which could lead to a reduction in the prevalence of HBV, HCV, syphilis, and HIV.

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