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A Prospective Study of Prevalence of Occult HBV Infection and Assessment of Risk Factors for HBV Transmission in Persons with Occult HBV Infection

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Occult HBV infection (OBI) is an important cause of cryptogenic cirrhosis and HBV reactivation during immunosuppression. There is limited data on OBI. Our aim was to study the prevalence of OBI in coastal Odisha and identify risk factors for HBV transmission in occult HBV infection. 173 consecutive apparently healthy volunteers were subjected to a questionnaire for risk factors for HBV infection. All underwent Anti-HBc assay, and those positive underwent PCR for HBV DNA. There were 173 HBsAg negative subjects; sex ratio was 7:1 [male:female] with mean age 39.2±10.6 years. Fifteen (8.7%) were anti-HBc positive, of whom 9(5.3%) were HBV DNA positive. Prevalence of risk factors in overall (171) and OBI (9) individuals were: history of jaundice (19/2; p =0.258), hospitalization(24/4; p =0.112), dental procedure (45/4; P =0.241), ear piercing (18/1; p =1.0), exposure to community barbers (39/5; p =0.028), promiscuity (15/3; P =0.032), childhood immunization (132/8; p =0.0688). On multivariate logistic regression analysis, exposure to community barbers (C I= 0.009-0.165, P =0.029) and promiscuity (CI= 0.032-0.263, P =0.013) were independent predictors of occult HBV infection. The prevalence of OBI in coastal Odisha was 5.3%. Exposure to community barbers and promiscuous sexual habit were independent predictors of occult HBV infection.

Keywords: Epidemiology, Hepatitis B, Occult HBV, Risk factors.

Hepatitis B is an infectious inflammatory illness of the liver caused by the hepatitis B virus (HBV) that affects hominoidea, including humans. Hepatitis B virus (HBV) remains a major public health problem worldwide.[1] According to World Health Organization estimate, two billion people worldwide have serologic evidence of past or present HBV infection, and 350 million are chronically infected and at risk for HBV related liver disease.[2] Approximately one third of all cases of cirrhosis and half of all cases of hepatocellular carcinoma can be attributed to chronic HBV infection. HBV is estimated to be responsible for 500,000–700,000 deaths each year.[2-4] A modest estimate would put the number of deaths occurring due to HBV infection per year in India to be around 150,000.[5]

In the era of molecular diagnostics, significant progress has been made in the understanding of the lifecycle, clinical course, pathogenesis, diagnosis and treatment of the hepatitis B virus (HBV).

HBsAg tests remain the first-line of blood screening for HBV. Current HBsAg screening assays are enzyme immunoassays (EIAs), including

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enzyme-linked immunosorbent assays and chemiluminescence immunoassays (CLIAs). These different assays have sensitivity ranging between <0.1 and 0.62 ng of HBsAg per mL (1ng/ml corresponds to approximately 2 IU/mL).[6,7] A chemiluminescent enzyme immunoassay (CLEIA) prototype has been developed that claims a sensitivity of 0.22 mIU/ml and the ability to reduce the window period by ~17 days compared to the CLIA systems in use.[8] Due to high cost and considerable equipment requirements, these HBsAg assays may not be affordable for small-scale blood services particularly in resource-limited developing countries. Rapid immunochromatographic HBsAg tests have been developed and evaluated in high prevalence areas.[9-11] Comparative studies showed that rapid tests are less sensitive than most EIAs.[9,10,12] Though HBV is transmitted by percutaneous or mucosal exposure to infected blood or other body fluids, transfusion-associated HBV continues to be a major problem in India, especially among multiple transfused patients despite testing for HBsAg in blood donors.[13] In India, detection of HBV infection among blood donors is carried out by HBsAg screening by commercial enzyme immunoassay. The prevalence of chronic HBV infection in India ranges from 2% to 10%, as shown by different studies.[14] Only one study on HBV infection has been reported from Orissa, which deals with the prevalence of HBsAg among the blood donors and reported 1.13% HBsAg positivity.[15] However, this strategy of screening of blood donors for HBsAg may sometimes fail in its purpose due to the presence of occult HBV infection. Occurrence of OBI in total anti-HBc positive subjects varies in different parts of India [0% in Chandigarh, 20.87% in New Delhi (Northern India), 30% in Ganjam and 21% in Kolkata].[13,16-19] Until recently, the clinical effect of OBI was unclear in the following contexts: its influence on the progression of liver disease or development of complications including hepatocellular carcinoma, and the risk for reactivation or transmission of HBV infection.[20] Thus in patients with OBI who are otherwise healthy, two key issues need resolution: development of long term complications due to HBV infection, and transmission pattern pertaining to occult HBV infection. There is very little data on these important issues. Also little is known about the risk factors of HBV infection in persons with OBI.

Aims and objectives

The focus of this study is on understanding the role of host factors in HBV transmission in coastal Odisha, especially with regard to OBI.

The main aims of this study are as follows:

1. To know the prevalence of OBI in the general population of coastal Odisha.
2. To study the association of different risk factors for transmission of HBV infection in persons with occult HBV infection

MATERIALS AND METHODS

This study was conducted during the period from September 2010 to December 2012. One hundred seventy three apparently healthy attendants accompanying gastroenterology patients were enrolled into the study with informed consent.

Inclusion Criteria

- Previously unscreened for HBV infection.
- Absence of overt liver disease.
- Good mental status.

Exclusion criteria

- HBsAg positive individuals.
- First degree relatives of HBV infected patients.
- Known case of chronic liver disease.

Sample Collection

Clinical data pertaining to HBV transmission for each individual were collected through appropriate questionnaire (Appendix 1). Three ml of blood sample was drawn from those individuals, who agreed for participation, after obtaining written consent.

Laboratory Procedures

Serological Assays

Commercially available enzyme linked immuno assay kits was used to test the serological markers of HBV infection. The HBsAg was detected by monoclonal antibody based test kits (Biomerieux, Boxtel, The Netherlands). anti-HBc (Hepanostika anti-HBc Uni-Form, Biomerieux, Boxtel, The Netherlands) was tested by commercial ELISA in HBsAg negative individuals. All the Anti-HBc positive samples were tested repeatedly and only those which were positive on repeated testing were considered as anti-HBc positive. Sera from the healthy attendants were stored at -80°C, and

were thawed once for serological examinations.

Serum HBV-DNA isolation and detection

The HBV DNA was extracted from 200 µl of serum by using standard viral DNA extraction kit.[21] Nested PCR was carried out using suitable primers.[22] HBV DNA amplification was done at least twice to rule out false positive results.

Assessment tools: Detailed history was taken from the volunteers selected for the study, and meticulous physical examination was performed. The data were recorded as per attached proforma.

Statistical analysis

All data were analysed using Statistical Package for Social Sciences (SPSS) version 17.0 and through online (http://statpages.org/ctab2x2.html) calculation, where necessary. The analysis was carried out at three levels of descriptive, bivariate and multivariate logistic regression analysis. Descriptive statistics of different characteristics of the sampled population were computed. Means and SD were calculated for quantitative variables and proportions for categorical variables. Percentage was used to describe the prevalence. OR and 95% CI was calculated for each association. Associations among independent variables were assessed using appropriate tests, such as x^2 or Fisher's exact tests, when indicated before performing logistic regression analysis. Multivariate logistic regression models were used to examine the association between independent variables and the main outcome variable, positive HBV DNA, while controlling for the effects of other covariates. All variables which were associated with outcome in bivariate analysis were included in the model. In logistic model, reference category (OR = 1) for OR estimate was HBV DNA negative participants. A probability of <0.05 was considered as statistically significant.

RESULTS

From September 2010 to December 2012, one hundred and seventy three apparently healthy attendants accompanying patients attending a gastroenterology clinic were enrolled in the study. In all of them HBs Ag was negative. 151(87.3%) were males and 22(12.7%) females, with a gender ratio of 7:1(M: F). Age range was between 12-71 years with the mean age being 39.2±10.6 years. Among them 15 (8.7%) were found to be total anti-HBc positive, of whom 9 (60%) were HBV DNA positive by nested PCR for surface region. Out of 151 males, anti-HBc was positive in 14(9.2%) males and 8(57.1%) of them were HBV DNA positive. Among 22 females, only one (4.5%) was total anti-HBc positive, and HBV DNA was detected in this female subject too (Fig. 1).

We evaluated prevalence of occult HBV infection along with the different risk factors in the healthy attendants (**Table 1**). None had present and/or previous history of ascites or upper gastrointestinal bleed. Nineteen (10.9%) patients had a prior history of jaundice with varying duration of 5-10 days. Two (10.5%) of them were found to be positive for total anti-HBc Ab, and HBV DNA too was detected in these individuals (100%).

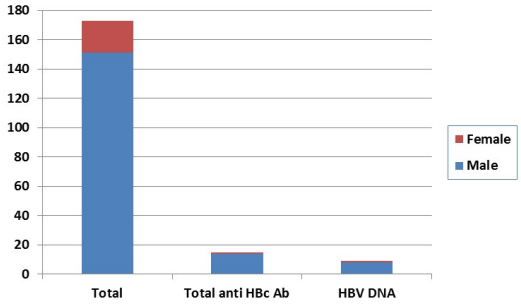

Fig. 1. Gender distribution of healthy attendants and occult HBV infection

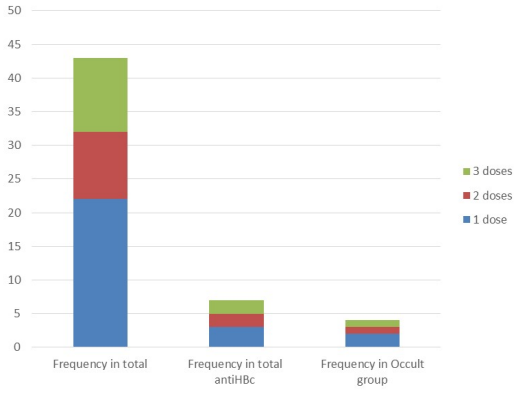

Fig. 2. Frequency of hepatitis B vaccine dose in healthy attendants and occult HBV infection

History of liver disease in family (alcoholic cirrhosis/death due to cirrhosis of unknown cause) was present in 11(6.3%) attendants but none of them was positive for total anti-HBc Ab. Further, none of family members of the attendants had hepatitis B infection. Twenty four (13.8%) subjects gave history of prior admission into hospital due to different medical and/or surgical problems. Out of the 24 subjects with history of prior hospitalizations, all were administered intravenous infusions, 8 had urinary catheterization, 17 had undergone surgery, 5 required blood transfusions, and 5 were administered repeated injections. Total anti-HBc Ab was present in 4(16.7%) attendants of whom three (75%) had HBV DNA. Among the catheterized 8 (4.6%), none was positive for total anti-HBc Ab. Out of 17(9.8%) persons with history of surgery, 2(11.7%) had total anti-HBc Ab, of whom one (50%) was HBV DNA positive. Out of the 5(2.9%) who had blood transfused, 2(40%) had anti-HBc Ab, of whom one (50%) was positive for HBV DNA. Similar result was also found in persons taking repeated injections. None of the volunteers had history of abscess drainage, circumcision, acupuncture. 20(11.5%) had done gastrointestinal endoscopy, in whom total anti-HBc Ab was present in 3(15%) persons and none had HBV DNA. History of dental procedure was found in 45(26%), in whom 9(20%) had positive anti-HBc Ab and 4(44.5%) had HBV DNA. 6(3.4%) volunteers had done tattooing, but only one(16.6%) had positive anti-HBc Ab without presence of HBV DNA. 18(10.4%,all females) had history of ear piercing in whom one(5.5%) had anti-HBc Ab with positive HBV DNA in same individual (100%) also. 51(29.5%) volunteers had donated blood, in whom 3(5.8%) had positive anti-HBc Ab and two(67%) had HBV DNA. Promiscuous sexual habits were found in 15(8.7%) volunteers, out of which anti-HBc Ab was present in 6(40%) and HBV DNA was present in 3(50%) volunteers. 39(22.5%) individual were exposed to community barbers. Positive anti-HBc Ab was present in 9(23%), in whom HBV DNA was found in 5(55.5%) persons. None of the study population had any history of local prevalence of hepatitis (HAV or HEV infection). Three (1.7%) volunteers had treated by village quacks, but none had positive anti-HBc Ab. Similarly three (1.7%) had history of vaccination for cholera, but none had positive anti-HBc Ab. 132(76.3%) individuals had taken childhood immunization under EPI. 14(10.6%) had positive anti-HBc Ab, among which 8(57.2%) had positive DNA. 43(24.8%) individuals had taken HBV vaccine, but 32(72%) individuals

Table 1. Frequency of study variables in different groups of study subjects

Variables	Total (n=173)	Total anti-HBcAb (n=15)	HBV DNA (n=9)
Age (years)	39.2±10.6	39.26±9.7	39.66±11.5
Male	151(87.3%)	14(9.27%)	8(57.1%)
Female	22(22.7%)	1(4.5%)	1(100%)
H/O Jaundice	19(10.9%)	2(10.5%)	2(100%)
Hospitalization	24(13.8%)	4(16.7%)	3(75%)
IV infusion	24(13.8%)	4(16.7%)	3(75%)
Surgery	17(9.8%)	2(11.7%)	1(50%)
Blood transfusion	5(2.9%)	2(40%)	1(50%)
Frequent injections	5(2.9%)	2(40%)	1(50%)
G. I. Endoscopy	20(11.5%)	3(15%)	0
Dental procedure	45(26%)	9(20%)	4(44.5%)
Tattooing	6(3.4%)	1(16.6%)	0
Ear piercing	18(10.4%)	1(5.5%)	1(100%)
Blood donation	51(29.5%)	3(5.8%)	2(67%)
Exposure to community barbers	39(22.5%)	9(23%)	5(55.5%)
Promiscuous sexual habit	15(8.7%)	6(40%)	3(50%)
Childhood immunization under EPI	132(76.3%)	14(10.6%)	8(57.2%)
HBV vaccination	43(24.8%)	7(16.3%)	4(57.2%)

had incomplete vaccination (single dose: 22, two doses: 10). In vaccinated group 7(16.3%) had total anti-HBc Ab and 4(57.2%) had HBV DNA of reactive anti-HBc population. Out of 22 single dose vaccine group, anti-HBc Ab was found in 3(13.6%) and of this HBV DNA was present in 2(67%). In double dose vaccinated individuals (10), anti-HBc Ab and HBV DNA were found in 2(20%) and 1(50.0%) respectively. Interestingly in 11(25.6%) completed vaccine group, total anti-HBc Ab was present in 2(18.2%) persons, from which 1(50%) had HBV DNA (**Figure 2**).

None have history of contact with HBV infected patients. There is no association of occult HBV infection between incomplete vaccine group and vaccinated (Odds ratio: 1.034, 95% CI: 0.077-29.032, p- value: 1.0).

Out of 9 occult HBV infection, 8(88.8%) were male. Only one female had occult HBV infection. Among all occult HBV infected patients, 2(22.2%) had past history of jaundice,3 (33.3%) were hospitalized(1: undergone surgery, 1: blood transfusion,1:history of multiple injections), 4(44.4%) had dental procedures, 5(55.5%) had exposure to community barbers, 4(44.5%) had promiscuous sexual activity, 1(11.1%)female had pierced the ear, 2 (22.2%) had donated blood, 4(44.5%) had taken hepatitis B vaccine, 8(88.8%) had history of childhood immunization under EPI. On bivariate analysis, exposure to community

Table 2. Bivariate analysis of risk factors in occult HBV infection

Variables	Total (n=173)	HBVDNA (n=9)	Odds ratio (95% CI)	p value
Gender (male)	151	8	1.175 (0.136-26.299)	1.0
H/O Jaundice	19	2	2.471(0.325-14.754)	0.258
Hospitalization	24	3	3.405 (0.618-17.109)	0.112
IV infusion	24	3	3.405 (0.618-17.109)	0.112
Surgery	17	1	1.156 (0.051-10.276)	1.0
Blood transfusion	5	1	5.0 (0.190-59.825)	0.237
Frequent injections	5 5	1	5.0 (0.190-59.825)	0.237
Dental procedure	45	4	2.40 (0.511-10.976)	0.241
Ear piercing	18	1	1.081(0.048-9.542)	1.0
Blood donation	51	2	0.671(0.093-3.711)	1.0
Exposure to community barbers	39	5	4.779 (1.038-22.709)	0.028
Promiscuous sexual habit	15	3	6.33 (1.087-34.339)	0.032
Childhood immunization under EPI	132	8	2.581(0.311-56.691)	0.688
HBV vaccination	43	5	4.145 (0.906-19.580)	0.043

Table 3. Comparison of prevalence of occult hepatitis B infection in India

Authors	Region/ Institution	Total subjects	Type of population	Anti-HBc reactive	Prevalence HBV DNA*
Chaudhuri V et al(2003)[13]	New Delhi	24,694[a]	Blood donors	3304(13.4%)[c]	27.2%(40/147)[e]
	6159[b]			230(3.7%)[d]	20.8%(48/230)[f]
Duseja A et al (2003)[16]	Chandigarh	100	Blood donors	0	0
Bhatacharya p et al (2007)[18]	Kolkata	1027	Blood donors	188(18.3%)	40(21.3%)
Asim M et al (2010)[26]	New Delhi	2175	Blood donors	413(19.8%)	31(7.5%)
Panigrahi R et al (2010)[19]	Ganjam	729	Blood donors	220(31%)	66(30%)
Present study	Cuttack	173	Healthy attendants	15(8.7%)	9(60%)

* Prevalence HBV DNA from total anti-HBc reactive

[a] Cohort 1, [b] Cohort 2, [c] Only anti-HBc results from cohort 1, [d] Only anti-HBc results from cohort 2, [e] Results from subset of only anti-HBc from cohort 1, [f] Results from subset of only anti-HBc from cohort 2.

barbers, promiscuous sexual habit and HBV vaccination were associated with occult HBV infection (**Table 2**), but on multiple logistic regression analysis exposure to community barbers (C I= 0.009-0.165, p=0.029), promiscuous sexual habit (C I= 0.032-0.263, p=0.013) were independent predictors of occult HBV infection.

DISCUSSION

Occult HBV infection has today emerged as a a global concern for all hepatologists, gastroenterologists, and other health care personnel especially public health experts, not only due to its association with cryptogenic liver disease and hepatocellular carcinoma, but also due to the disease burden in the general population. Surprisingly, despite this anxiety, we know very little about the transmission pattern of HBV in this setting of seemingly healthy general population. Most of the prevalence studies of Occult HBV infection were performed in blood donors. Further, there is no information on transmission pattern of occult HBV infection from India. Several reports from around the world have revealed detection of HBV DNA in blood of HBsAg-negative individuals and hence suggested transmission of HBV infection by blood transfusion from HBsAg negative occult HBV carriers[23,24] It is believed that evaluation of other HBV markers might reduce this risk.

In present study, we have enrolled healthy volunteers accompanying gastroenterology patients attending an outpatient clinic; this would result in a homogenous distribution of the study population from different regions of Odisha. In this study total anti-HBc Ab was present in 8.7% of attendants, but HBV DNA was present in only 60% of the total anti-HBc Ab positive subjects. Thus HBV DNA was present in 5.3% of the general population from coastal Odisha. Earlier studies have demonstrated that HBV DNA was detected in 3% to 38% of blood donors with those who are negative for hepatitis B surface antigen (HbsAg) and hepatitis B surface antibody (HBs Ab) but positive for total anti-HBc antibody.[19,25] Varying prevalence of antiHBc, a marker for exposure to HBV infection, has been reported from different parts of India - ranging between 2% to 31% of total donor population (Table 3 and 4). Dhawan et al tested 1700 samples, and anti-HBc reactive was found in 142 (8.4%) blood samples. Of them HBV DNA was detected in only 1 of 100 samples tested.[17] A donor with a positive total anti HBc-IgG indicates either a past infection or a carrier state. Anti-HBc IgG may remain positive for life in an affected individual although the individual has protective levels of anti-HBs and therefore, this does not necessarily mean that the blood of such a donor is infectious. However, unless we do sensitive NAT we can't confirm the presence or absence of the virus in such donor. Unlike IgG subtype, anti-HBc IgM is a marker of recent hepatitis B infection, which was taken account into in two recent studies.[29,30] Kumar et al[29] in a blood bank based study observed that total of 11 out of 2552 (0.43%) blood units were reactive for HBcAg IgM, and 112 out of 704 (15.9%) samples were reactive for total anti-HBcAg. In another study by Shastry et al,[30] on screening for HBcAb IgM in 12,232 healthy voluntary blood donors, 15(0.1%) were found to be positive, although none was HBsAg reactive.

Table 4. Comparison of prevalence of occult hepatitis B infection with other countries.

Authors	Region/ Institution	Total subjects	Type of population	anti-HBc reactive	Prevalence HBV DNA*
Yotsuyanagi H et al (2002)[31]	Japan	Unknown	Blood donors	50	19(38%)
Kleinman SH et al(2003)[32]	North America	395	Blood donors	107(27%)	4(3.7%)
S.Ramia et al (2005)[33]	Lebanon	2505	Blood donors	54(2.2%)	7(12.9%)
Behzad- Behbahani et al (2006)[27]	Iran	2000	Blood donors	131(6.55%)	16(12.2%)
Bhatti FA et al (2007)[28]	Pakistan	966	Blood donors	167(17.28%)	5(2.9%)
Present study	Cuttack	173	Healthy attendants	15(8.7%)	9(60%)

* Prevalence HBV DNA from total anti-HBc reactive

Further, the frequency of occult HBV infection varies considerably from different parts of the world, and this depends on the prevalence of HBV infection in the study population. Prevalence of OBI is high in high HBV prevalence zone of the world and low in low HBV prevalence zone of the world. Studies from different parts of India **(Table 3)** have reported occult HBV infection ranging from 21% in Kolkata (Eastern India), and 20.87% in New Delhi (Northern India) to 0% in Chandigarh (Northwestern India).[16-18]

A study from Japan[31] has reported DNA positivity of 38% (19 of 50) in anti-HBc reactive samples. In contrast, a study from North America found 3.7%, HBV DNA positivity among 107 anti-HBc positive/anti-HBs negative samples **(Table 4)**.[32] In a recent study from Berhampur,[19] Ganjam in southern Odisha, about 30.1 % (220/729) of total donations were total antiHBc positive, and 30%(66/220) of the positive samples had HBV DNA. This has been attributed to more promiscuous habits due to high migration and more number of HIV positive people in the district of Ganjam. In contrast, the present study from a different part of the same state of Odisha shows a lower Anti-HBc positivity, but a greater HBV DNA positivity (60%) among the total anti-HBc positive subjects, highlighting the stark regional variations.

Besides, there is increasing evidence that OBI is associated with chronic liver disease and HCC,[34,35] in addition to being a source of transmission of HBV by blood transfusion or orthotropic liver transplantation.[36] Therefore, the high rate of OBI among healthy attendants is of serious concern as this population has the potential to transmit HBV by transfusion of contaminated blood through the public blood supply and other modes of transmission, since they are negative on routine screening for HBsAg. Thus, our data indicates OBI as an emerging infection hindering safety of blood transfusion and increasing cause of HBV endemicity in the community. Ideally, HBsAg negative individuals who are either antiHBc negative or those who are antiHBc positive/HBV DNA negative should be allowed as regular blood donors to minimize transmission due to occult hepatitis B infection. This will help in the reduction of the transfusion associated transmission due to OBI, but will increase the screening costs. Routine blood donor screening for anti-HBc has been implemented in some countries resulting in a decrease in the risk of post-transfusion HBV infection.[32]Hence in our resource poor setting too, inclusion of anti-HBc testing for donor screening will definitely remove possible HBV infected donations. Thus although a large number of donations will be rejected, these rejections of anti-HBc positive donations will be valuable in reducing the risk of HBV transmission with its potential consequences, especially among immune compromised recipients.

Current population of India in 2016 is 1,312,716,811 (1.31 billion) with a total male Population 677,886,961 (677 million)and total female population 634,829,849 (634 million). Presuming an overall anti-HBc Ab prevalence rate of 3-31%, this can be translated to 39 to 407 million populations with reactive anti-HBc Ab. So because of screening a large population of India will be deprived of donating of blood in resource poor country like India. This is definitely a huge burden and cannot be ignored.

From analysis of the different risk factors, exposure to community barbers and promiscuous sexual habit were strongly associated with occult HBV infection. In many developing countries like India, most people shave their beards and moustache and get their hair cut by the community barbers with common razors. Besides, there is absence of awareness amongst the barbers about the HBV infection and its modes of transmission. Similarly lack of awareness regarding HBV infection and its pattern of transmission especially among sex workers may also be a factor for occult HBV infection.

Further, those with incomplete vaccination had more occult HBV infection. However, one subject with three doses of vaccine was also found to have occult HBV infection, which might be explained by either prior infection before vaccination or mutant HBsAg due to vaccine pressure. The vast majority of the general population do not have awareness about HBV vaccination, as only 24.8% of the population had been vaccinated with HBV vaccine, with only 6.3% having completed three doses of vaccine. A study by Misra *et al.,* from Odisha had earlier found that only one third of the population were aware about hepatitis B and its vaccine, and less than a third of the population were vaccinated for hepatitis B.[37]

Another study from Mumbai in 2002 highlighted the lack of awareness even among medical and nursing students. Besides, this study also revealed that only 26.3% of the medical students had taken 3 doses of hepatitis B vaccination.[38] Another study from Lahore, Pakistan to assess the vaccination status among HCWs and medical students also found that only 49% health care workers and 42.20% medical students were vaccinated.[39]

CONCLUSIONS

The present study showed that the prevalence of OBI in coastal Odisha is as high as 5.3%. This raises major safety concerns regarding blood donation by apparently healthy persons after they are screened only for HBsAg. Since, exclusion of anti-HBc Ab positive units from donor pool is not practical in areas with intermediate HBs Ag prevalence such as India, and discarding anti HBc positive units would result in unacceptably high rates of donor rejection, the usefulness of screening for anti-HBc Ab as an additional screening test to improve the safety of the blood supply in India deserves further studies. Besides, exposure to community barbers and promiscuous sexual habit were found to be strongly associated with OBI; however, the possible role of other risk factors for transmission of OBI requires larger population based studies. Further, measures to increase awareness regarding modes of HBV transmission among health care personnel, the general public, barbers and sex workers are necessary to reduce the burden of occult HBV infection in India.

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