

HBsAg Carriers among Healthy Nepalese Men: A Serological Survey

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ABSTRACT

The prevalence of hepatitis B surface antigen (HBsAg) was studied among 2,585 healthy Nepalese men, aged 16-50 years, who required medical check-ups for employment abroad. Serum samples, collected during July-September 1999, were tested for HBsAg using a third-generation ELISA kit. Of the 2,585 men, 24 (0.93%) were positive for HBsAg, indicating that hepatitis B infection in the target group was below the intermediate endemicity. The prevalence of HBsAg was minimum (0.36%) in the central development region, followed by western, eastern and mid-western development regions (0.82%, 1.16%, and 2.08% respectively, $\chi^2=4.76$, $p<0.2$). The positivity of HBsAg was slightly higher in the hilly region (1.11%) than in the terai (0.84%) and mountainous regions (0%), which was not significant ($\chi^2=2.1$, $p<0.5$). The prevalence was highest in the 46-50-year age group. Alanine aminotransferase was tested in all 24 positive and 150 negative subjects to indicate the stage of infection. Seven (29.16%) of the 24 positive cases had elevated alanine aminotransferase, indicating associated liver damage ($\chi^2=4.16$, $p<0.05$) and it was significantly associated with positivity of HBsAg ($\chi^2=32.6$, $p<0.001$). All 9 positive subjects from the terai region had normal alanine aminotransferase, whereas 7 of the 15 subjects from the hilly region had it elevated ($\chi^2=11.76$, $p<0.001$). Seven (29%) of the positive subjects were chronic carriers with its sequel in the liver, and the remaining 17 subjects (70.84%) may be in the incubation period or convalescent stage or may be chronic carriers. The results suggest that the population should be educated about the sequel of the infection.

Key words: Hepatitis; Hepatitis B virus; HBsAg; Serodiagnosis; Epidemiology; Alanine aminotransferase; Disease transmission; Nepal

INTRODUCTION

Nepal has three different regions: terai (0-300 m above the sea-level), hilly (600-3,000 m above the sea-level), and mountainous (3,000 m above the sea-level). Administratively, the country is divided into eastern, central, western, mid-western, and far-western development regions.

At the time when one-third population (2 billion) of the world has been infected with hepatitis B virus (HBV) and about 350 million people in the world are chronic

carriers of HBV (1), ordinary Nepalese population, especially those in rural area, are not aware of its existence and sequel of its infection.

Ninety-eight percent of babies born to mothers with chronic HBV infection become infected, and 95% of them develop persistent infection (2). By contrast, 90% of adults infected with HBV clear their acute infection and become immune. Nine percent of them become chronic carriers, and these carriers are the reservoir of HBV. One percent of chronic carriers suffer from fulminant hepatic failure and lead to jaundice, hepatocellular carcinoma, or liver cirrhosis.

HBV is the most common cause of chronic liver disease and hepatocellular carcinoma in Asian countries, except Japan (3), and is an important cause of morbidity and mortality. In a previous study in Nepal, the 3.97% seroprevalence of HBsAg among Nepalese healthy male

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population was reported to belong to the area of intermediate endemicity (4) on the basis of the categorization made by Zuckerman and Harrison in 1990 (5). The present study was aimed at finding the prevalence of the HBsAg carriers among healthy male population by development regions in Nepal.

MATERIALS AND METHODS

The subjects of the study included healthy males from different regions of Nepal who required medical check-ups for employment abroad and exhibited no symptom of liver inflammation. They came mostly from rural areas lacking any knowledge on HBV infection, its transmission and sequel, and most of them were unaware of safe sex. The male population was selected because they are exposed to various kinds of risk factors of HBV infection.

Blood samples collected from 2,585 healthy males aged 16-50 years were tested on the same day for serological detection of hepatitis B surface antigen (HBsAg), using the third-generation ELISA HBsAg kit

regions. The sample size from the far-western development region was very small, hence it was not included in data analysis. Statistically, no association was observed between the regions and the occurrence of HBV infection ($\chi^2=4.76$, $p<0.2$).

The positivity of HBsAg was high in the hilly region (1.11%) compared to the terai (0.84%) and mountainous regions (0%) (Table 2).

The association between HBV infection and hilly region was not significant ($\chi^2=2.1057$, $p<0.5$). The

Table 1. Prevalence of HBsAg in different development regions

Region	No. of samples tested	HBsAg-positive	% of prevalence
Eastern	1,036	12	1.16
Central	550	2	0.36
Western	851	7	0.82
Mid-western	144	3	2.08
Far-western	4	-	-
Total	2,585	24	0.93

Table 2. Positivity of HBsAg in different regions

Region	Eastern			Central			Western			Mid-western			Total	%
	Total	No.	%	Total	No.	%	Total	No.	%	Total	No.	%		
Terai	542	7	1.29	348	1	0.28	136	1	0.73	39	0	0	1,065	0.84
Hilly	390	5	1.28	185	1	0.54	671	6	0.89	105	3	2.85	1,351	1.11
Mountainous	104	0	0	17	0	0	44	0	0	-	-	-	165	0
Total	1,036	12	1.15	550	2	0.36	851	7	0.82	144	3	2.08	2,581	0.93

(double-antibody sandwich) (Bioelisa, Biokit, Barcelona, Spain). The kit had a sensitivity of 0.125 HBsAg unit per mL for 'ad' and 'ay' subtypes. Standard methodology as specified by the manufacturer was followed. The absorbance of wells (hence the positivity of samples, validity of test, and cut-off value) was measured at 450 nm using ELISA Reader ELX 800G (USA). One hundred fifty negative subjects and all the samples positive for HBsAg were tested for determination of alanine aminotransferase as an index of liver damage, using SGPT enzymatic kit (Ranbaxy, India) and auto-analyzer Biotron 810 (USA). Chi-square test was used for analyzing data.

RESULTS

Twenty-four (0.93%) of the 2,585 subjects were positive for HBsAg, indicating active HBV infection. The maximum (2.08%) occurrence was observed in the mid-western development region (Table 1), followed by the eastern (1.16%) and western (0.82%) development

greater prevalence in the hilly area, compared to the terai area, was observed in the mid-western development region (2.85% vs 0%) but was not statistically significant ($\chi^2=1.176$, $p<0.5$). No significant difference in the prevalence was observed in the eastern, central and western development regions.

HBV infection was observed to be 2.14% (5 in 233), 1.26% (11 in 973), 1.13% (1 in 88), and 1.03% (4 in 388) in the age groups of 36-40, 21-25, 41-45 and 31-35 years respectively. The prevalence was minimum (0.15%, 1 in 652) in the age group of 26-30 years, followed by the age group of 16-20 years (0.5%, 1 in 201). The maximum prevalence (5.56%, 1 in 18) was observed in the 46-50-year age group. However, no association between HBV infection and age was observed ($\chi^2=9.41$, $p<0.1$). The age groups of 46-50 and 41-45 years were not included in statistical analysis due to a small sample size, and ages of 2,453 subjects were known.

In 24 HBsAg-positive and 150 HBsAg-negative subjects, alanine aminotransferase was performed by the enzymatic method in which any value above 49 IU/L was considered abnormal. Alanine aminotransferase was observed high in 7 of 24 positive subjects (29.16%), indicating the damage of liver, the sequel of HBV infection. This elevated alanine aminotransferase had significant association with positivity of HBsAg ($\chi^2=4.16$, $p<0.05$). Of the 150 HBsAg-negative subjects, only 2 (1.33%) had elevated alanine aminotransferase, suggesting the association of positivity of HBsAg with elevated alanine aminotransferase, indicating the damage in liver ($\chi^2=32.66$, $p<0.001$).

Of the 24 positive subjects, all nine from the terai area had normal alanine aminotransferase, whereas 7 of the 15 subjects of the hilly region had elevated alanine aminotransferase (Table 3), indicating the association

and also not consistent with the report of Manandhar and Shrestha. Furthermore, Sawayama *et al.* had reported the prevalence of 1.1% HBV infection in Bhadrakali and Kolyang villages of Nepal (8). In different Nepalese studies, the higher prevalence of HBV infection has been reported among hospital patients [5% (9), 8.8% (10)], sex workers [10.9% (11)], and also among general population of certain districts [6.6% (10)]. Of the blood donors in Nepal, 1.26% were positive for HBsAg (12) which is almost consistent with the finding of the present study and the observation of Shrestha (6,7). The seroprevalence of HBV infection obtained in the target group was low compared to 4% of HBV infection in India (13) and also was low compared to stated 20% of chronic carriers in many developing countries (14).

The high positivity of HBsAg in the hilly region (1.11%) compared to the terai region (0.84%) and in the

Table 3. Relation of alanine aminotransferase with HBsAg carriers from hilly region

Region	Alanine aminotransferase					
	Terai			Hilly		
	Normal	High	Total	Normal	High	Total
Eastern (12)	7	0	7	2	3	5
Central (2)	1	0	1	1	0	1
Western (7)	1	0	1	5	1	6
Mid-western (3)	0	0	0	0	3	3
Total	9	0	9	8	7	15

of positivity of HBsAg in them with elevated alanine aminotransferase ($\chi^2=11.766$, $p<0.001$).

DISCUSSION

In the present study, 0.93% of the subjects had HBV infection, showing the endemicity of HBV below intermediate (2-7%) on the basis of categorization by Zuckerman and Harrison (5). This finding is consistent with the observation of Shrestha who reported that about 1% of the total Nepalese population was infected by HBV (6,7), but not with the finding of Manandhar and Shrestha who reported the prevalence (in 478 subjects) of 3.97% (4) that included subjects from all development regions. The difference between the findings of these two studies could be due to a very small sample size in the present study from the far-western development region where a maximum prevalence (6.2%) was reported by Manandhar and Shrestha. In the present study, although the prevalence was high (2.08%) in the mid-western development region and minimum (0.36%) in the central development region where capital city Kathmandu is located, it was not statistically significant

mountainous region (0%), along with the findings of various researchers, suggests that variations in prevalence can occur in different districts of the same development region. The cause of dissimilarity between the findings of Manandhar and Shrestha and those of the present study could be due to this factor and the sample size.

Although HBsAg marker is the most useful marker in detecting acute and chronic infections, detection of HBsAg alone cannot ascertain the stage of infection, since it is also positive in different other stages. The test for all markers, such as anti-HBc, anti-HBsAg, HBeAg, and anti-HBe, to identify the stage of infection of each positive subject was not possible in this study. Since the study subjects were healthy with no symptoms of icterus, fatigue, nausea, etc., they could be chronically infected with HBV. Therefore, the alanine aminotransferase value could show evidence of liver damage, and it was significantly high in 29.16% of the positive subjects, indicating the damage to liver caused by HBV infection. Similarly, Joshi has reported elevated alanine aminotransferase in 36.15% of HBsAg-positive subjects

among blood donors (12). It was also elevated significantly in subjects from the hilly region, and the cause of the association was not understood. However, it could be due to the immune response of population, climatic condition, or dietary habit of subjects in the present study. Over 29% of the positive cases with elevated alanine aminotransferase indicate that they were chronic/persistent carriers of HBV with indication of liver damage, and its significant association with the subjects of hilly region indicates that the population from the hilly region should be the priority group for vaccination and educational programmes. The remaining 70.84% of the positive cases may have infection at the incubation or prodromal period or at the convalescence stage, or were persistent carriers. Asymptomatic patients with no history of acute hepatitis and normal liver function may also be chronic carriers.

Since the target group of the present study was those seeking medical check-ups for employment abroad, older and younger age group could not be included despite the susceptibility of paediatric age group toward HBV infection. Although the prevalence was not alarmingly high, it is time to educate Nepalese people about HBV infection, its transmission, and the consequences.

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