The Prevalence of Occult Hepatitis B Virus in the Hemodialysis Patients in Yazd, Iran

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Abstract- Occult HBV infection of hemodialysis (HD) patients is informative in terms of virus transmission. It may be of clinical importance in HD patients. The aim of this study was to investigate the prevalence of anti-HBc in the HD Patients. Number of 126 patients undergoing hemodialysis were included in this study from main hemodialysis units in Yazd. Hepatitis B surface antigens (HBsAg), hepatitis B core antibody (anti-HBc) were examined in all subjects. Finally, stored serum samples from anti-HBcAb positive, HBsAg negative patients were anonymised and tested for HBV DNA by real time quantitative PCR assay. The age range of the patients was 17-88 years. Of the 126 patients, 123 patients (97.6%) were HBC-Ab negative and 3 (2.4%) were positive. Of 3 patients with Anti-HBC positive, HBV DNA was detected in 1 patient. This study showed a low rate of isolated anti-HBc (2.4%). HBV DNA was also detected in 1 patient.
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Introduction

Occult HBV infection (OBI) is the persistence of viral genome in the liver tissue in individuals negative for HBsAg. OBI is defined by the presence of HBV DNA in the liver (with detectable or undetectable HBV DNA in the serum) in patients with serological markers of previous infection (anti-HBc and/or anti-HBs positive) or in patients without serological markers (anti-HBc and/or anti-HBs negative) (1). HBV affects all age groups and can lead to liver disease, liver cancer, and death in many of those afflicted (2-4).

The use of hemodialysis (HD) for end-stage renal disease (ESRD) has increasingly expanded in the past decades (5,6). Hemodialysis patients are at risk of acquiring parenterally transmitted infections such as HBV, because of the large number of receiving blood transfusions, invasive procedures they undergo, shared dialysis equipment, impaired host immune response and lower response rates to HBV vaccination (6,7). This virus may be transmitted through shared-use dialysis machinery, or via frequent blood transfusions or by reactivation after kidney transplantation, leading to the progression of liver disease (6,7). In general, about 20% of individuals with HBV have negative results for all HBV serological markers (seronegative group), and 80% have positive results for serological markers of previous infection with HBV (seropositive group). In total, 35% of patients with HBV have positive results for anti-HBs and 42% of them show anti-HBc positivity. The important issues of occult hepatitis B are the aggravation of chronic liver damage and concomitant fibrosis (7,8).

HBV infection is most frequently seen in patients with anti-HBc as the only HBV serological marker (isolated anti-HBc), and the HBV-DNA detection rate is higher in subjects who are anti-HBc positive but anti-HBs negative (7,8). Iran is a low endemic area of HBV infection (8). For example, the prevalence of HBV in the

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hemodialysis patients in the Khuzestan Province has been reported to be 5.1%. It seems that the prevalence of hepatitis virus infections is higher in developing countries than in developed countries (9,10).

Strict adherence to HBV infection control measures has led to a decline in HBV prevalence in hemodialysis units (10-15). These measures include; routine screening of donated blood products, separation of HBV dedicated dialysis machines, HBV vaccination and periodic measurement of anti-HBV antibodies (15-17).

This study was carried out to determine the prevalence of anti-HBc in the hemodialysis Patients in the Yazd, Iran.

Materials and Methods

The study population comprised 126 patients undergoing hemodialysis at The Hospitals of Yazd, Iran in 2014-2015. HBsAg negative patients were selected and enrolled inthestudy. Demographic data on age, gender and during the period of hemodialysis were collected from the renal database.

Sampling for this study was performed for patients undergoing hemodialysis. After describing the purpose of research for patients, sampling was done from the brachial area on the elbow by the laboratory personnel of the dialysis part. Then, serum and plasma were separated and were referred to the virology part of the lab.

The presence of anti-HBc was determined by ELISA (Enzyme-Linked Immunosorbent Assay) test (Diasorin, Italy). DNA was extracted from patient's materials using the commercial Kit (Roche, Germany) according to manufacturers instructions. The extracted DNA was stored at -20° C till used.

The plasma sample is processed with a starting material of 200 µ and eluted in 50 µ of elution buffer by a spin procedure. Twenty microliters of elutewere used as a template for PCR amplification with the artus HBV RG PCR kit (Qiagen, Germany) on the Rotor-Gene 6000 platform (Corbett Research, Mortlake, Victoria, Australia). The assay targets a 134-bp region of the HBV core gene, and the detection limit determined by the manufacturer is 20 IU/ml (95% detection limit).

Statistical analysis

Data analysis was performed using the Statistical Package for the Social Sciences 16.0 (SPSS Inc., Chicago, IL, USA). $P<0.05$ was accepted as significant.

Results

In this study, 126 hemodialysis patients were presented. All the demographic information and their disease have been shown in table 1.

Toinvestigate,HBC-Ab was usednon-Competitive Sandwich ELISA method. Of the 126 patients, 123 patients (97.6%) were HBC-Ab negative and 3 (2.4%) were positive (Table 2).

To detect HBV DNA in the patients of HBC-Ab negative was performed Real-time PCR. Of 3 patients with Anti-HBC positive, HBV DNA was detected in 1 patient. This person was a diabetic 45-year-old man that had high blood pressure with more than a 5-year history of hemodialysis period. Also, the level of its education was the level of literacy.

Table 1. The demographic information of patients

Parameters			N	%
Age	Under 40 years		12	9.5
	40-60 years		53	42.1
	Above 60 years		61	48.4
Gender	Male		82	65.1
	Female		44	34.9
Level of education	Illiterate		35	27.8
	Reading literacy		44	34.9
	Below diploma		15	11.9
	Diploma		22	17.5
	Associate degree and above		10	7.9
During the period of hemodialysis	Under 2 years		37	29.3
	2-5 years		49	38.9
	Above 5 years		40	31.7
Disease	High blood pressure	With Dis.	45	35.7
		Without Dis.	81	64.3
	Diabetes mellitus	With Dis.	63	50
		Without Dis.	63	50

The age range of the patients was 17-88 years.

Table 2. HBC-Ab test results and its frequency based on demographic data and disease

Parameters			HBC-Ab				*P*.value
			Negative		Positive		
			N	%	N	%	
Age	Under 40 years		10	100	0	0	0.821
	40-60 years		49	98	1	2	
	Above 60 years		64	97	2	3	
Gender	Male		79	96.4	3	3.7	0.190
	Female		44	100	0	0	
Level of education	Illiterate		34	97.1	1	2.9	0.730
	Reading literacy		43	97.7	1	2.3	
	Below diploma		14	93.3	1	6.7	
	Diploma		22	100	0	0	
	Associate degree and above		10	100	0	0	
During the period of hemodialysis	Under 2 years		37	100	0	0	0.037
	2-5 years		49	100	0	0	
	Above 5 years		37	92.5	3	7.5	
Disease	High blood pressure	With Dis.	43	95.6	2	4.4	0.251
		Without Dis.	80	98.8	1	1.2	
	Diabetes mellitus	With Dis.	61	96.8	2	3.2	0.550
		Without Dis.	62	98.4	1	1.6	

Discussion

The present study was aimed to investigate and determine the prevalence of anti-HBc in the hemodialysis Patients in the Yazd, Iran. Of 3 patients with Anti-HBC positive, HBV DNA was detected in 1 patient. 123 patients were HBC-Abnegative, and 3 were positive.

A study was carried out to detect hepatitis B virus among hemodialysis patients in Khartoum State, Sudan.Out of the patients sampled, 65 were males, and 35 were females (age 18 to 70 years) none of these patients showed signs of clinical hepatitis. The results showed that 9 out of the 100 samples were positive for HBsAg, and were subsequently excluded from the study. Out of the remaining HBsAg negative 91 samples, 38(51.6%) showed positive HBc antibodies and 3 (3.3%) tested positive for HBV DNA using competitive ELISA and PCR, respectively (18). In present study also of 3 patients with Anti-HBC positive, HBV DNA was detected in 1 patient.

Another study was performed to investigate the prevalence of hepatitis B virus in a cohort of Egyptian patients maintained on hemodialysis. Twenty-five individuals were HBV DNA-positive, representing 26.8% of the tested patients. Three patients (12%) were positive to both anti-HBc and anti-HBs. Finally, fifteen patients (60%) were positive to anti-HBc only (19). This study also was similar to our study, but positive cases for anti-HBc, and HBV DNA was higher towards present study.

Another study was conducted to determine the prevalence of hepatitis B virus among all HBsAg negative hemodialysis patients. A hundred HBs Ag negative HD patients with a median age of 60 years were enrolled in this study. Finally, isolated anti-HBc was detected in 2% of cases. HBV DNA was detected in 1% of HBsAg-negative patients (20). This study was similar tothepresent study and positive cases for HBV DNA, and anti-HBc were a low number.

The low rate of isolated anti-HBc (2.4%) can be due totheimprovement of knowledge about HBV transmission routes, HBV vaccination among hemodialysis patients. In addition, investigating hemodialysis patients for anti-HBc can show evidence of latent infection in them.

References

1. Su T, Li C, Wang J, Chen Q, Feng Y, Shi J, et al. Study on risk factors of hepatitis B virus infection among patients receiving hemodialysis by multi-level statistical model analysis. Zhonghua Liu Xing Bing XueZaZhi 2015;36:510-4.
2. Sowole L, Labbett W, Patel M, O'Riordan A, Cross J, Davenport A, et al. The prevalence of occult hepatitis B virus (HBV) infection in a large multi-ethnic haemodialysis cohort. BMC Nephrol 2015;16:12.
3. Gasim GI, Bella A, Adam I. Immune response to hepatitis B vaccine among patients on hemodialysis. World J

Hepatol 2015;7:270-5.
4. Yari A, Yousefzadeh H, Tahaghoghi S, Ghazvini K. Prevalence of Hepatitis B and C among Patients Looking for Hospital Care; Five Years' Study in Mashhad, Iran.Acta Med Iran 2016;54:54-7.
5. Li X, Kang H, Wang S, Deng Z, Yang T, Jia Y, et al. Knowledge, attitude, and behavior of hepatitis B virus infection among chinese dental interns. Hepat Mon 2015;15:25079.
6. Saijo T, Joki N, Inishi Y, Muto M, Saijo M, Hase H. Occult hepatitis B virus infection in hemodialysis patients in Japan. TherApher Dial 2015;19:125-30.
7. Eleftheriadis T, Pissas G, Antoniadi G, Liakopoulos V, Stefanidis I. Factors affecting effectiveness of vaccination against hepatitis B virus in hemodialysis patients. World J Gastroenterol 2014;20:12018-25.
8. Kim SM, Kim HW, Lee JE, Lee EK, Shin HD, Song IH. Occult hepatitis B virus infection of hemodialysis patients: a cross-sectional study in a hepatitis B virus-endemic region. HemodialInt 2015;19:66-71.
9. Bokharaei-Salim F, Keyvani H, Monavari SH, Esghaei M, Fakhim S, AtaeiPirkooh A, et al. Distribution of hepatitis B virus genotypes in azerbaijani patients with chronic hepatitis B infection. Hepat Mon 2014;14:e25105.
10. Keyvani H, Sohrabi M, Zamani F, Poustchi H, Ashrafi H, Saeedian F, et al. A population based study on hepatitis B virus in northern iran, amol. Hepat Mon 2014;14:20540.
11. Nagakawa O, Miyatomi Y, Shigeta Y, Inayama E, Murakami K, Sakai T, et al. Importance of isolated anti-hbc in detection of occult hepatitis B virus infection in hemodialysis patients: reply. TherApher Dial 2013;17:644-5.
12. Spaziante M, Biliotti E, Grieco S, Palazzo D, Esvan R, Taliani G. Anti-HBs seroconversion during treatment with entecavir in a patient with chronic hepatitis B virus infection on hemodialysis. J Med Virol 2014;86:139-43.
13. Yassin MH, Gupta V. Role of infection control in prevention of hepatitis B virus (HBV) in hemodialysis (HD) patients. Infect Disord Drug Targets 2013;13:162-8.
14. Rinonce HT, Yano Y, Utsumi T, Heriyanto DS, Anggorowati N, Widasari DI, et al. Hepatitis B and C virus infection among hemodialysis patients in Yogyakarta, Indonesia: Prevalence and molecular evidence for nosocomial transmission. J Med Virol 2013;85:1348-61.
15. Fontenele AM, Filho NS, Ferreira AS. Occult hepatitis B in patients on hemodialysis: a review. Ann Hepatol 2013;12:527-31.
16. Einollahi B, Alavian SM. Hepatitis B virus infection: need for more attention in hemodialysis patients. Saudi J Kidney Dis Transpl 2013;24:587-8.
17. Ramezani A, Banifazl M, Eslamifar A, Ahmadi F, Razeghi E, Aghakhani A. Outcome of hemodialysis patients with occult hepatitis B virus infection at a 4 year follow-up. J Gastrointestin Liver Dis 2013;22:111-2.
18. Mohammed AA, Enan KA, Khair OM, Hussien MO, El Hussein ARM, Elkhidir IM. Prevalence of occult hepatitis B virus (HBV) infections in haemodialysis patients in Khartoum State, Sudan from 2012 to 2014. J Med Lab Diagn 2015;6:22-6.
19. Elgohry I, Elbanna A, Hashad D. Occult hepatitis B virus infection in a cohort of Egyptian chronic hemodialysis patients. Clin Lab 2012;58:1057-61.
20. Ramezani A, Aghasadeghi MR, Ahmadi F, Razeghi E, Eslamifar A, Banifazl M, et al. Isolated anti-hbc and occult HBV infection in dialysis patients. Nephrourol Mon 2014;7:e22674.