

Evaluation of hepatitis B virus and Hepatitis C virus frequency in hemodialysis patients

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Abstract

Aim: This study aimed to investigate the prevalence of the hepatitis B virus and hepatitis C virus in patients undergoing hemodialysis in Karabuk city.

Material and Methods: For patients presented to the Karabuk University Training and Research Hospital for hemodialysis between January 2016 and December 2018, HBsAg, anti-HBs, and anti-HCV assays were conducted using the chemiluminescence method and HBV DNA, HCV RNA and HCV genotyping tests were conducted using real-time polymerase chain reaction. The data was retrospectively evaluated.

Results: Of 345 patients undergoing hemodialysis, 211 (61.2%) were male and 134 (38.8%) were female. The mean age of the patients was 62 (15–96) years. Moreover, 0.9% (n = 3) of patients were HBsAg positive and 4.3% (n = 15) were anti-HCV positive. Three patients with HBsAg positivity were also HBV DNA positive. Of the 15 patients who were anti-HCV positive, 12 were also HCV RNA positive. Of these, eight patients had the HCV genotype 1b and four patients had the genotype 4. Anti-HBs seropositivity was detected in 65.8% of patients. The highest Anti-HBs seropositivity rate was observed in patients aged <40 years (76%) and the difference between age groups was statistically significant (p = 0.002).

Conclusion: In our study, the prevalence of HBsAg seropositivity in patients undergoing hemodialysis (0.9%) was lower than that reported by the Turkish Society of Nephrology (2.65%). This difference may arise from the high anti-HBs seropositivity rate. Conversely, the prevalence of anti-HCV seropositivity (4.3%) was similar to the mean prevalence in Turkey (4.2%). Infection prevention measures against viral hepatitis should be strictly implemented, especially in hemodialysis units.

Keywords: Hemodialysis; hepatitis B; hepatitis C

INTRODUCTION

Currently, viral hepatitis is a global public health concern. Annually, it accounts for 1.4 million deaths worldwide, of which 90% are caused by the hepatitis B virus (HBV) and hepatitis C virus (HCV) (1,2). Viral hepatitis increases mortality and morbidity in patients undergoing hemodialysis (HD) owing to cellular immunodeficiency induced by uremia, frequent blood transfusions, and long-term exposure to infectious agents through the extracorporeal circulation (3,4). The incidence of HBV infection among patients undergoing HD has gradually decreased with the introduction of hepatitis B vaccines in the 1980s and after healthcare service providers started to use separate hemodialysis devices for patients with

hepatitis B (5-8). Conversely, a significant reduction in the incidence of HCV infections has not been achieved as an HCV vaccine has not been developed yet. The prevalence of HCV among patients undergoing HD is <5% in high-income countries and 4%–60% globally (9,10).

This cross-sectional study aimed to investigate HBV DNA, HCV RNA, and HCV genotypes as well as HBsAg, anti-HBs, and anti-HCV seropositivity in patients undergoing HD at the Karabuk University Training and Research Hospital.

MATERIAL and METHODS

HBsAg, anti-HBs, anti-HCV, HCV RNA, HBV DNA, and HCV genotyping results of 345 patients undergoing HD who presented to the Karabuk University Training and

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Research Hospital between January 2016 and December 2018 were included in this cross-sectional study. Duplicate results were excluded from the study. First, the serologic parameters were evaluated for each patient. Patients were then divided into three groups based on age: <40 years, 41–65 years, and >65 years. As the study was retrospectively designed, test results were obtained from the database of the microbiology laboratory. Ethic approval for the study was obtained from the Non-Interventional Research Ethics Board of the Karabuk University (decision dated 04/02/2019 and numbered 2/29).

HBsAg, anti-HBs, and anti-HCV tests were conducted on serum samples sent to the microbiology laboratory using the ARCHITECT i2000 SR (Abbott Diagnostics, USA) device using the chemiluminescence method. The HCV RNA test was performed on anti-HCV positive samples. HBV DNA was assayed in HBsAg positive samples. First, nucleic acid isolation was performed for HBV DNA and HCV RNA using the Magnesia 16 (Anatolia Geneworks, Turkey) device. Then, the quantitative HBV DNA and HCV

RNA tests were conducted using the Montania 4896 (Anatolia Geneworks, Turkey) device using real-time polymerase chain reaction (RT-PCR) in accordance with the manufacturer's instructions. The HCV genotyping test was conducted using the Montania 4896 device by RT-PCR using the Bosphore HCV genotyping (Anatolia Geneworks, Turkey) kit that can detect four major and most predominant HCV genotypes (1, 1a, 1b, 2, 3, 4) of HCV in patients with HCV RNA positivity.

Statistical analysis

The Statistical Package for the Social Sciences, version 25.0, (Armonk, NY, USA) software was used for data analysis. The Kolmogorov–Smirnov test was used to determine whether the data was normally distributed. Descriptive statistics were expressed as numbers, percentages, and median values (minimum, maximum). The Pearson's Chi-square test was used for analyzing groups, and $p < 0.05$ was considered statistically significant.

Table 1. Distribution of HBsAg, anti-HBs and anti-HCV seropositivity according to age and gender in patients undergoing hemodialysis

	HBsAg		anti-HBs		anti-HCV	
	Positive n (%)	Negative n (%)	Positive n (%)	Negative n (%)	Positive n (%)	Negative n (%)
Gender						
Female	1 (0.7)	133 (99.3)	92 (69)	42 (31)	2 (0.2)	132 (98.8)
Male	2 (0.9)	209 (99.1)	135 (64)	76 (36)	13 (6.1)	198 (93.9)
Age groups						
<40 years	-	29 (100)	22 (76)	7 (24)	-	29 (100)
41-65 years	1 (0.6)	158 (99.4)	117 (74)	42 (26)	10 (6.2)	149 (93.8)
>65 years	2 (1)	155 (99)	88 (56)	67 (44)	5 (3)	152 (97)
Total	3 (0.9)	342 (99.1)	227 (65.8)	118 (34.2)	15 (4.3)	330 (95.7)

Table 2. Age and gender information and HCV molecular test results of anti-HCV positive patients

Patient ID	Gender	Age	anti-HCV (S/CO)	HCV RNA (IU/ml)	HCV genotype
14	M*	66	15.6	3890	1b
16	M	55	12.46	Negative	-
17	M	55	13	332500	1b
46	M	55	14.7	422300	4
90	F**	53	15.9	1.102.000	1b
98	F	83	8.85	Negative	-
124	M	49	15.6	3350	1b
133	M	61	15.43	790300	4
140	M	64	9.21	422300	4
148	M	76	13.85	542700	1b
172	M	53	12.73	995900	4
206	M	74	12.91	Negative	-
258	M	60	10.7	1334	1b
261	M	58	14.11	4119000	1b
271	M	73	17.3	960600	1b

*: Male ** :Female

RESULTS

Of 345 patients with HD, 211 (61.2%) were male and 134 (38.8%) were female. The mean age of the patients was 62 (15–96) years. Moreover, 65.8% of patients ($n = 227$) had anti-HBs seropositivity, which had a similar prevalence in males and females.

Seropositivity rate was 76% for patients aged <40 years and 56% for those aged >65 years; this difference was statistically significant ($p = 0.002$). HBsAg positivity and anti-HCV seropositivity rates were 0.9% ($n = 3$) and 4.3% ($n = 15$), respectively. Of 15 patients who were anti-HCV-positive, 13 were male and two were female (Table 1).

Three of these patients were HCV RNA-negative. Of the 12 patients who were HCV RNA-positive, eight had the genotype 1b and four had the genotype 4 (Table 2). Three HBsAg positive samples were also HBV DNA positive. None of the patients had both HBsAg and anti-HCV seropositive.

DISCUSSION

Viral hepatitis B remains an important health concern both in Turkey and worldwide. According to the 2017 data by the World Health Organization, there were approximately 257 million HBV carriers worldwide, of which 887,000 patients will die because of cirrhosis or hepatocellular carcinoma (11). Turkey is among the moderately endemic countries with a hepatitis B seropositivity rate of 2%–7% (12,13). In this study, we found that HBsAg seropositivity rate was 0.95% among patients undergoing HD and living in Karabuk. The Turkish Society of Nephrology reported that the prevalence of HBsAg seropositivity in patients undergoing HD in Turkey was 6.8% in 2015, 3.3% in 2016, and 2.9% in 2017 (14). Moreover, a study by Karlıdag et al. (15) and Kızılates et al. (16) determined that the prevalence of HBsAg positivity in patients undergoing HD was 5.8% and 2.4%, respectively. Such low prevalence of HBsAg in Turkey can be attributed to the high anti-HBs seropositivity rate. In the present study, anti-HBs seropositivity rate was 65.8% for all groups; moreover, the highest anti-HBs seropositivity rate was observed in patients aged <40 years (76%) and the lowest rate in patients aged >65 years (56%) ($p = 0.02$). This may be associated with the widespread use of HBV vaccines and a lack of sufficient immune response to HBV vaccination in patients aged >65 years who have chronic kidney failure and who have been undergoing HD for a long time (9). In the literature, the prevalence of anti-HBs seropositivity among patients undergoing HD was found to be 56.1% by Guimarães et al. (17) and an even higher rate of 88.7% by Savcı et al. (18).

The risk of HCV infection is higher in patients undergoing HD owing to frequent blood transfusions and dialysis. There are currently more than 71 million patients with HCV infection worldwide and 399,000 patients die each year due to HCV-related cirrhosis or liver cancer (19). However, a vaccine that is effective against HCV has not

yet been developed owing to the high genetic variability of HCV. Therefore, the prevention of HCV infections is of paramount importance (20). According to the results of a research conducted by the Turkish Association for the Study of the Liver, anti-HCV seropositivity rate was 0.95% in Turkey (21). Moreover, HCV carriers undergoing HD were associated with a 1.57 fold increased risk of death (22). In the present study, 4.3% of patients undergoing HD had anti-HCV positivity and 3.4% of those had HCV RNA positivity. According to the 2017 report of the Turkish Society of Nephrology, 4.2% and 2.9% of patients undergoing HD in Turkey had anti-HCV and HCV RNA seropositivity, respectively (14). Similarly, Temiz et al. found that anti-HCV positivity rate was 4.1% in patients undergoing HD (7). Conversely, Karlıdag et al. found a low anti-HCV positivity rate of 1.2% among patients undergoing HD. They linked this to the effective implementation of infection control measures as well as a small number of blood transfusions due to the administration of erythropoietin in these patients (15).

In addition to the HCV RNA assay, identifying the HCV genotype in patients with anti-HCV positivity is essential to determine the treatment protocol and duration. HCV genotypes 1, 4, 5, and 6 are more resistant to therapy and require a long treatment duration (10). In this study, of 12 patients with HCV RNA positivity, eight had the genotype 1b and four had the genotype 4. Genotype 1b is particularly more common in America, Europe, Turkey, and Russia (23,24), whereas genotype 4 is more common in North Africa and in Middle Eastern countries (25). Although genotype 1b is more common in Turkey, the prevalence of other genotypes, particularly genotypes 3 and 4, has also started to increase in recent years (26). We believe that this is influenced by the recent intense migration movements observed in Turkey. Reportedly, the prevalence of genotype 4 was higher especially among Syrian refugees (25). Our study also showed the presence of genotype 4 in four patients in addition to genotype 1 in the patients and all of them were Turkish citizens. Considering the relationship between HCV genotypes and age, it was reported that patients with genotype 1b or 2 were older than those with genotypes 1a, 3 and 4 (27). In our study, all patients who had the genotype 4 and 1b aged <65 and >65 years, respectively.

This is the first study investigating the prevalence of HBV, HCV, and anti-HBs among patients undergoing HD and living in Karabuk. Results revealed that 0.9% of patients had HBsAg positivity and 4.3% of patients had anti-HCV seropositivity. The most commonly detected genotype in patients with HCV RNA positivity was genotype 1b, followed by genotype 4. In addition, anti-HBs positivity rate was 65.8%.

The HBsAg positivity is used to diagnose of Hepatitis B infection but only anti-HBc IgG can be positive which is usually indicative of past HBV infection in 10–20% of patients (28). In our hospital, anti-HBc IgG test is not

routinely requested by clinicians. Therefore isolated anti-HBc IgG positivity and vaccination status of patients could not be determined. This is the limitation of our study.

CONCLUSION

In conclusion, the strict implementation of the necessary infection control measures is required to protect patients undergoing HD, who have a considerable risk of blood-borne infections from viral hepatitis. Blood transfusions and invasive procedures should be limited in these patients. Moreover, both patients and medical staff should be provided with periodic trainings to increase awareness regarding blood-borne infections.

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REFERENCES

1. Jefferies M, Rauff B, Rashid H, et al. Update on global epidemiology of viral hepatitis and preventive strategies. *World J Clin Cases* 2018;6:589-99.
2. Kayar Y, Dertli R. Factors affecting hepatitis B immunization in celiac disease. *Ann Med Res* 2019;26:1478-83.
3. Kamal IMA, Mahdi BM. Seroprevalence occurrence of viral hepatitis and HIV among hemodialysis patients. *Ann Med Surg* 2018;29:1-4.
4. Roushan MRH, Farokhtabar S, Bayani M, et al. Epidemiological Aspects of Hepatitis B and C and Human Immunodeficiency Viruses Among Hemodialysis Patients in Mazandaran Province, Iran. *Nephrourol Mon* 2016;8:37878.
5. Demirbakan H, Daglar D, Ergani A, et al. Investigation of Hepatitis B and Hepatitis C Virus Infections by Serological and Molecular Methods in Hemodialysis Patients. *Mikrobiyol Bul* 2014;48:143-50.
6. Ayub MA, Bacci MR, Fonseca FLA, et al. Hemodialysis and hepatitis B vaccination: a challenge to physicians. *Int J Gen Med* 2014;7:109-14.
7. Temiz H, Kaya S, Berekatoglu N, et al. The Evaluation of the Seroprevalence of HBV, HCV and HIV Infections and the Assessment of Antibody Response to Hepatitis B Vaccination in Hemodialysis Patients. *Viral Hepat J* 2013;19:140-3.
8. Cordeiro VM, Martins BCT, Teles SA, et al. Decline in hepatitis B and C prevalence among hemodialysis patients in Tocantins. Northern Brazil. *Rev Inst Med trop S Paulo* 2018;60:36.
9. Fabrizi F. Hepatitis C virus infection and dialysis: 2012 update. *ISRN Nephrol* 2013;2013:159760.
10. Ozer Etik D, Ocal S, Boyacioglu AS. Hepatitis C infection in hemodialysis patients: A review. *World J Hepatol* 2015;7:885-95.
11. Hepatitis B. World Health Organisation (WHO). <https://www.who.int/news-room/fact-sheets/detail/hepatitis-b> access date 18 July 2019.
12. Akarca US. Chronic hepatitis B. A guideline to diagnosis, approach, management, and follow-up 2007. Turkish Association for the Study of Liver. *Turk J Gastroenterol* 2008;19:207-30.
13. Ege B, Kucuk AOA, Koparal M. Investigation preoperative seroprevalence of HBsAg, anti-HBs, anti-HCV, anti-HIV and vaccination level of patients undergoing dental surgical procedure under local anesthesia. *Ann Med Res* 2019;26:1503-7.
14. Seyahi N. Türk Nefroloji Derneği 2017 yılı Türk Böbrek Kayıt Sistemi. <http://www.nefroloji.org.tr/folders/file/TND-2017-Kayit-Sistemi-Verileri.pdf> access date 20 July 2019.
15. Eser Karlidag G, Kuçuksu M, Demir M. HBsAg, Anti-HBs, Anti-HCV and HIV Seroprevalence in Hemodialysis Patients in Elazığ Province. *Viral Hepat J* 2018;24:53-6.
16. Kizilates F, Berk H, Coban M, et al. Seroprevalence of hepatitis B and C virus in patients who undergo hemodialysis in Antalya province, Turkey. *Asian Biomed* 2016;10:339-44.
17. Guimaraes MNC, Facincani T, Santos SSD. Hepatitis B status in hemodialysis patients. *Arq Gastroenterol* 2017;54:356-8.
18. Savcı U, Bulut Y. Polymerase chain reaction with investigation of occult Hepatitis B infection in hemodialysis patients. *Turk J Clin Lab* 2015;6:19-23.
19. Hepatitis C. World Health Organization (WHO). <https://www.who.int/news-room/fact-sheets/detail/hepatitis-c> access date 18 July 2019.
20. Odemis I, Kose S, Tatar B, et al. Hepatitis A, B, C and HIV seroprevalence among young health care workers: A cross-sectional study. *Behcet Uz Cocuk Hast Derg* 2018;8:8-14.
21. Tozun N, Ozdogan OC, Cakaloglu Y, et al. A Nationwide Prevalence Study and Risk Factors for Hepatitis A, B, C and D Infections in Turkey. Boston: AASLD National Meeting in Boston; 2010. p. A789.
22. Fabrizi F, Martin P, Dixit V, et al. Meta-analysis: Effect of hepatitis C virus infection on mortality in dialysis. *Aliment Pharmacol Ther* 2004;20:1271-7.
23. Selcuk H, Kanbay M, Korkmaz M, et al. Distribution of HCV genotypes in patients with end-stage renal disease according to type of dialysis treatment. *Dig Dis Sci* 2006;51:1420-5.
24. Perez RM, Ferraz ML, Figueiredo MS, et al. Unexpected distribution of hepatitis C virus genotypes in patients on hemodialysis and kidney transplant recipients. *J Med Virol* 2003;69:489-94.
25. Abdel-Ghaffar TY, Sira MM, El Naghi S. Hepatitis C genotype 4: The past, present, and future. *World J Hepatol* 2015;7:2792-810.
26. Kirisci O, Caliskan A. The Predominance of Genotype 3 in Hepatitis C Virus in the Province of Kahramanmaraş,

- Turkey/Genotype Distribution of Syrian Refugee Patients with Hepatitis C in Kahramanmaras Province. Jundishapur J Microbiol. 2019;12:80872.
27. Mohamed NA, Zainol RZ, Wong KK, et al. Hepatitis C genotype and associated risks factors of patients at University Kebangsaan Malaysia Medical Centre. Pak J Med Sci 2013;29:1142-6.
28. Jain M, Chakravarti A, Kar P. Clinical significance of isolated anti-hbc positivity in cases of chronic liver disease in new delhi, India. J Glob Infect Dis 2009;1:29-32.