Iranian Journal of Virology 2014;8(4): 25-30
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Original Article

Diagnostic and Epidemiologic Features of Hepatitis C among Hemodialysis Patients Living in Yazd Province of Iran

Baghbanian M[1]*, Karimi-Bondarabadi AA[2], Doosti M[3], Shayestehpour M[4], Salmanroghani H[1], Keyvani H[5], Dastamouz S[2], Ahmadi A[6], Moallemi Z[7], Shahidokht M[8]

1. Department of Gastroenterology, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
2. Research Center of Hematology & Oncology and Genetics, Department of Internal Medicine, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
3. Department of Virology, Faculty of Medical Sciences, Tarbiat Modares University, Tehran, Iran.
4. Department of Virology, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran.
5. Department of Virology, Iran University of Medical Sciences, Tehran, Iran.
6. Department of Microbiology, Golestan University of Medical Sciences, Gorgan, Iran.
7. Dr Infection and tropical disease. Infection and tropical disease research center Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
8. Department of pathology Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Abstract

Background and Aims: Hepatitis C is a blood-borne virus that causes a liver inflammation. The prevalence of hepatitis C (HCV) in hemodialysis patients is more than the general population. Periodic screening of HCV in dialysis centers is necessary to control this viral disease. The aim of this study is the evaluation of HCV infection among hemodialysis patients living in Yazd province of Iran.

Materials and Methods: In this cross sectional study, two milliliters blood sample has been taken from each of 188 hemodialysis patients. Third-generation enzyme-linked immunosorbent assay (ELISA) and RT-PCR methods were carried out for detecting HCV antibody and viral genome in patient sera. Demographic and clinical characteristics of patients were collected and data were analyzed by Fisher's exact test and Mann–Whitney U test in SPSS software v.19.

Results: HCV-Ab was positive in 12 hemodialysis patients**.** Eight cases had a history of blood transfusion and four patients had a history of kidney transplantation. Anyone didn't have a history of skin tattooing or intravenous drug usage. HCV-RNA was positive in 4 cases and was negative in 8 patients with positive HCV-Ab.

Conclusion: HCV prevalence using ELISA and RT-PCR tests among hemodialysis patients living in Yazd provenance of Iran is 6.3% and 2.1%, respectively. Risk factors are the hemodialysis duration ($p<0.001$) and kidney transplantation ($p=0.005$).

Keywords: Enzyme-Linked Immunosorbent Assay, Hepatitis C, Renal Dialysis

Introduction

Liver disease is a significant cause of morbidity and mortality in patients with end-stage renal disease (ESRD) treated by dialysis or kidney transplantation. Hepatitis C virus (HCV) infection is the most common liver disease in renal dialysis patients (1). Prevention, diagnosis and treatment of Hepatitis C among hemodialysis patients is problematic (2).

The world health organization (WHO) reported that about 3% of global population suffering hepatitis C (3). The prevalence of this infection in hemodialysis patients is more than the

***Corresponding author:** Mahmud Baghbanian, MD; Gastrointestinal Endoscopy Ward, Shaheed Sadoughi Hospital, Ebne sina Square, Yazd, Iran. Tel: (+98) 351-8224004, Fax: (+98) 351-8224100
Email: baghbanian1352@gmail.com

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general population. Anti HCV in hemodialysis patients is reported 11.6% in the United States to 91% in some developing countries (4, 5). The worldwide prevalence of HCV infection in hemodialysis patients approved with reverse transcriptase polymerase chain reaction (RT-PCR) is 5-24% (6, 7).

Risk factors are known for HCV in hemodialysis patients, including intravenous drug injection, past blood transfusion, kidney transplantation, long duration of dialysis, and tattoo (7, 8). The routine tests for HCV detection in patients are enzyme-linked immunosorbent assay (ELISA) and RT-PCR. Serological assays for anti-HCV may underestimate the frequency of HCV infection in this immunocompromised population. Virology assays are necessary for accurate diagnosis (9, 10). HCV-Ab for diagnosing hepatitis C infection has some defects, because dialysis patients are immunosuppresed cases and HCV-Ab may be undetectable despite active HCV infection. False-positive results are possible in patients with autoimmune disease such as systemic lupus erythmatous that may be associated with renal failure. The serology test is positive for a long time in persons who have improved from HCV (11). HCV-RNA PCR has high sensitivity and specificity, and it is the best method for HCV assay.

Screening for HCV in Iran is initiated since 1996. Several reports showed that the prevalence of HCV is less than 1% in Iranian general population (12). In previous studies, HCV seroprevalence (HCV antibody positivity) among Iranian hemodialysis patients has reported 5.5% to 55.9% (13, 14). However, because HCV-Ab test is not confirmative, the true prevalence of HCV infection is not clear in these studies. We don't encounter to these high rates of positive HCV serology among our hemodialysis patients in the center of Iran. In 2006, Iran Hepatitis Network (IHN) and Iran Ministry of Health and Medical Education started a project for control of HCV in dialysis patients (15). In 2011, Samimi-Rad et al. reported the prevalence of HCV antibody among hemodialysis patients 5.0% in Yazd province (16), but further study in this HD center is necessary. Periodic screening of Hepatitis C virus in dialysis units is necessary for control this viral disease. We can have the dialysis units without HCV infection if we follow the standard infection precautions and check these centers for HCV infection periodically. The aim of this study is the evaluation of HCV infection by serological (ELISA) and molecular (RT-PCR) methods among hemodialysis patients living in Yazd province of Iran.

Methods

This cross-sectional study is approved by Yazd Shahid Sadoughi University of medical sciences ethical committee and is conducted at 2012-2014 in Shahid Sadoughi hospital that is the capital hemodialysis center of Yazd province. All of 188 patients who were on hemodialysis in this center enrolled in the study. The inclusion criteria were all patients of any age, any sex, dialysis duration more than one month and residence in Yazd provenance. Exclusion criterion was immigration to Yazd provenance lees than one years ago. Patients gave informed consent to participate in study. Two milliliters venous blood sample has been taken from each patient. The plasma samples were been isolated and were frozen in -70°C. Then HCV-Ab and qualitative HCV-RNA was measured in each sample. Third-generation enzyme-linked immunosorbent assay (ELISA) was used for testing HCV-Ab. Our method for testing qualitative HCV-RNA was the polymerase chain reaction (RT-PCR). RNA extraction was carried out using high pure viral nucleic acid kit (Roch, Germany). The important variants that were assessed as a risk factor for HCV infection were intra venous drug injection, past blood transfusion, kidney transplantation, duration of dialysis, and tattoo. Demographic and clinical characteristics of patients were collected, and descriptive results were reported as mean±SD. Data analyzed by Fisher's exact test and Mann–Whitney U test in SPSS software v.19.

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Results

One hundred and eighty-eight patients were participated in study. One hundred and twenty six cases were males, and sixty two patients were females. The average age of them was 57±14.3 years old (range 23 to 87). HCV-Ab is tested before beginning of the hemodialysis and annually for each patient in our center. HCV-Ab was positive in one case at the beginning of the renal hemodialysis; however, HCV-RNA was negative in this case.

HCV-Ab was positive in 12 patients (6.38%), average age of them was 50.8±17.6, six cases were male, and average dialysis duration was 97±83 months. Etiology of renal failure was diabetic nephropathy in four cases, lupus nephropathy in four patients, polycystic kidney disease in two case, and nephrolithiasis in two patient. Ten cases had history of blood transfusion, and four cases had history of kidney transplantation. Anyone of these 12 patients didn't have history of intravenous drug usage or skin tattooing (Table 1).

HCV-RNA was positive in four cases (2.1%). Two cases were 81 years-old man with polycystic kidney disease that were on hemodialysis for 40 months. They didn't have history of blood transfusion, kidney transplantation, and intravenous drug usage or skin tattooing. Two other cases were 54 and 60 years-old women with diabetic nephropathy and rejected kidney transplant that were on hemodialysis for 156 months.

There wasn't any patient with negative HCV-Ab and positive HCV-RNA. HCV-RNA was negative in 8 patients with positive HCV-Ab. All of four patients who were HCV-RNA positive had positive HCV-Ab. The prevalence of PCR approved HCV infection in our patients was 2.1% that is four folds more than general population in Iran. Risk factors related to these subjects included in Table 1. We assessed HCV infection risk factors such as intra venous drug injection, history of blood transfusion, kidney transplantation, duration of dialysis, and tattoo. Our study showed significant relation between HCV infection with hemodialysis duration ($p<0.001$) and history of kidney transplantation ($p=0.005$).

Discussion

Prevention, diagnosis, and treatment of hepatitis C in dialysis patients is challenging and problematic. This study has determined the diagnosis and prevalence of HCV infection among hemodialysis patients living in Yazd

Table 1: Demographic and clinical characteristics of screened patients.

Variables	Positive HCV markers group (total=12)	Negative HCV markers group (total=176)	*P* values
Age (years)	50.83±17.57	57.43±14.14	0.345
Gender Male	6	120	0.392
Female	6	56	
Dialysis duration (months)	97.0±83.0	37.90±29.16	p<0.001
Blood transfusion (n)	8	80	0.316
Kidney transplantation (*n*)	4	8	0.005
IV Drug abuser (n)	0	2	0.79
Tattoo (n)	0	4	0.71

provenance of Iran by serology (HCV-Ab) and molecular virology (RT- PCR) methods. The prevalence of HCV Ab positivity was 6.38% in our study that is six fold more than Iranian general population. Ansar MM et al. reported 55% HCV-Ab positivity in hemodialysis patients living in north of Iran (14). This high rate of HCV seroprevalence positivity may be against the true in this domain, because hemodialysis patients are relatively immunocompromised, and we expect these patients exhibit HCV-Ab fewer than non-hemodialysis patients. This overt difference between our study and Ansar MM may be due to false positivity of HCV-Ab test related to low accuracy of serologic method or true positive test after HCV infection clearance in Ansar MM et al. study. This high prevalence of HCV-Ab positivity in Ansar MM study doesn't indicate true prevalence of HCV infection, because is not approved by molecular method (RT- PCR).

Mohtasham Amiri et al. reported that HCV prevalence in hemodialysis patients of Gilan province was 8.24% (17). The previous studies in other provinces and cities of Iran reported that HCV prevalence in these patients is similar to our results. Prevalence of Hepatitis C in hemodialysis patients in Jahroom city, Markazi, Tabriz, Isfahan and Khuzestan was 5.9%, 5.4%, 8.1%, 5.2% and 7.9%, respectively (18).

Prevalence of HCV-Ab positivity (6.4%) is more than HCV RNA PCR positivity (2.1%) in our study. The HCV antibody test is very sensitive and can sometimes cross-react with other antibodies. False-positive ELISA tests are not uncommon. This condition can be seen in patients who have cleared the hepatitis C virus after acute infection or by therapy because of remaining antibodies. This unexpected finding may be related to underlying autoimmune/rheumatologic disorders that lead to false positivity of HCV-Ab. In low-risk population such as blood donors, the false positivity of ELISA has been recorded up to 25% (19). There is also always the possibility of lab error leading to a false-positive test result. We evaluated the HCV infection risk factors such as intra venous drug injection, history of blood transfusion, kidney transplantation, duration of dialysis, and tattoo. Our study showed significant relation between positive HCV markers with hemodialysis duration ($p<0.001$) similar to Somi et al. study in Tabriz(8), Amiri et al. study in Gilan (17) and Bozorgi study in Qazvin. Hosseini-Moghadam et al. were not found a relation between mean duration of hemodialysis therapy with HCV infection in Tehran city (20). In our study, HCV infection had not been significant relation with intra venous drug injection ($p=0.79$), blood transfusion ($p=0.316$), and tattoo ($p=0.71$). Somi et al. were reported a relationship between renal transplantation with HCV infections (8), but we not founded any significant relation.

Hemodialysis (HD) is known as an important cause of viral hepatitis infections (7, 20). The high prevalence of HCV among dialysis patients can be related to several risk factors including the number of blood transfusions, dialysis duration and type of dialysis (lower HCV prevalence among peritoneal dialysis patients). Dialysis unit Equipment's and personnel's have important role in HCV transmission. Sharing of a multi-dose heparin vial between patients and unchanging gloves during hemodialysis treatment for new patient are as the causes of transmission of the infection in dialysis centers. Several study are reported the incidence of HCV infection in dialysis patients who shared dialysis machines in dialysis unit (21).

Despite the performance of common health care such as cleaning and disinfection of hemodialysis membranes and sterilizing all the equipment, which is relevant to HD patients, the special surveillance of staff, general rules for asepsis, virological testing with high assurance should be practically common in the dialysis centers in Iran (22). Some studies are reported a prevalence of HCV-Ab in HD patients 50% in Saudi Arabia, 20% in Turkey, and 21% in Jordan (23) Our study shows an acceptable HCV infection seroprevalence (6.38%) Vs. China (16.7%), America (8.9%), France (16.3%) and Egypt (80%) (24). Prevalence of HCV infection in HD patients substantially has improved in Iran from 14.4%

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in 1999 to 4.5 in 2006 (25). In 2011, Samimi-Rad et al. reported the prevalence of HCV among HD patients 5.0% in Yazd province while in our study, this amount was 2.1%. Therefore, the rate of HCV infection in HD centers can decrease if follow the standard infection precautions.

HCV prevalence using ELISA and RT-PCR tests among hemodialysis patients living in Yazd provenance of Iran is 6.3% and 2.1%, respectively. Risk factors are hemodialysis duration ($p<0.001$) and kidney transplantation ($p=0.005$).

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